

EAST Search History

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L1	171	cysk or cysteine synthase\$1	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 13:07
(L2)	127	1 same (gene\$1 or sequence\$1 or nucleic or polynucleotide\$)	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:03
L3	118438	(serine or ser) same (coexpress\$ or rich or high or level\$1 or yield\$1 or optimiz\$)	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:05
L4	63	1 and 3	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:06
(L5)	18	4 not 2	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:15
L6	6745	(amino acid\$1) near5 protein\$1 near5 (production\$ or express\$ or optimiz\$)	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:19
L7	949	6 same coli	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:18
L9	206	(amino acid\$1) near5 (composition\$ or profile\$) near5 protein\$1 near5 (production\$ or express\$ or optimiz\$)	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:24
(L10)	5	9 same coli	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:21
(L11)	12	(amino acid\$1) near5 (composition\$ or profile\$) near5 (heterologous or foreign) near3 protein\$1 near5 (production\$ or express\$ or optimiz\$)	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:27
(L12)	1	1 same coexpress\$	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:28
(L13)	77	1 same (serine or ser)	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:29
(L14)	20	13 not 2	US-PGPUB; USPAT	ADJ	OFF	2006/03/10 16:29

* * * * * * * * * * * * * * * * STN Columbus * * * * * * * * * * * * * * *

FILE 'HOME' ENTERED AT 17:09:38 ON 10 MAR 2006

=> fil .bec
COST IN U.S. DOLLARS
FULL ESTIMATED COST

| SINCE ENTRY | TOTAL SESSION |
|-------------|---------------|
| 0.21 | 0.21 |

FILES 'MEDLINE, SCISEARCH, LIFESCI, BIOTECHDS, BIOSIS, EMBASE, HCAPLUS, NTIS,
ESBIOBASE, BIOTECHNO, WPIDS' ENTERED AT 17:09:59 ON 10 MAR 2006
ALL COPYRIGHTS AND RESTRICTIONS APPLY. SEE HELP USAGETERMS FOR DETAILS.

11 FILES IN THE FILE LIST

=> s cysk or cysteine synthase#
FILE 'MEDLINE'

77 CYSK
64899 CYSTEINE
92418 SYNTHASE#
228 CYSTEINE SYNTHASE#
(CYSTEINE (W) SYNTHASE#)
L1 265 CYSK OR CYSTEINE SYNTHASE#

FILE 'SCISEARCH'

52 CYSK
47434 CYSTEINE
107943 SYNTHASE#
200 CYSTEINE SYNTHASE#
(CYSTEINE (W) SYNTHASE#)
L2 233 CYSK OR CYSTEINE SYNTHASE#

FILE 'LIFESCI'

48 CYSK
18083 "CYSTEINE"
23974 SYNTHASE#
88 CYSTEINE SYNTHASE#
("CYSTEINE" (W) SYNTHASE#)
L3 120 CYSK OR CYSTEINE SYNTHASE#

FILE 'BIOTECHDS'

51 CYSK
4197 CYSTEINE
6065 SYNTHASE#
59 CYSTEINE SYNTHASE#
(CYSTEINE (W) SYNTHASE#)
L4 79 CYSK OR CYSTEINE SYNTHASE#

FILE 'BIOSIS'

76 CYSK
59077 CYSTEINE
99656 SYNTHASE#
217 CYSTEINE SYNTHASE#
(CYSTEINE (W) SYNTHASE#)
L5 273 CYSK OR CYSTEINE SYNTHASE#

FILE 'EMBASE'

58 CYSK
49436 "CYSTEINE"
90470 SYNTHASE#
194 CYSTEINE SYNTHASE#
("CYSTEINE" (W) SYNTHASE#)
L6 223 CYSK OR CYSTEINE SYNTHASE#

FILE 'HCAPLUS'

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    175 CYSK
100024 CYSTEINE
  94834 SYNTHASE#
   359 CYSTEINE SYNTHASE#
      (CYSTEINE(W) SYNTHASE#)
L7      451 CYSK OR CYSTEINE SYNTHASE#

FILE 'NTIS'
    0 CYSK
  490 CYSTEINE
  232 SYNTHASE#
   0 CYSTEINE SYNTHASE#
      (CYSTEINE(W) SYNTHASE#)
L8      0 CYSK OR CYSTEINE SYNTHASE#

FILE 'ESBIOBASE'
    41 CYSK
 23741 CYSTEINE
 44267 SYNTHASE#
   96 CYSTEINE SYNTHASE#
      (CYSTEINE(W) SYNTHASE#)
L9      122 CYSK OR CYSTEINE SYNTHASE#

FILE 'BIOTECHNO'
    43 CYSK
 22339 CYSTEINE
 29457 SYNTHASE#
   130 CYSTEINE SYNTHASE#
      (CYSTEINE(W) SYNTHASE#)
L10     151 CYSK OR CYSTEINE SYNTHASE#

FILE 'WPIDS'
    44 CYSK
  8434 CYSTEINE
  4985 SYNTHASE#
   43 CYSTEINE SYNTHASE#
      (CYSTEINE(W) SYNTHASE#)
L11     58 CYSK OR CYSTEINE SYNTHASE#

TOTAL FOR ALL FILES
L12     1975 CYSK OR CYSTEINE SYNTHASE#

=> s (serine or ser) (15a) (rich or high or level# or yield# or optimiz?)
FILE 'MEDLINE'
    89374 SERINE
   21253 SER
   82402 RICH
 1384133 HIGH
 1482581 LEVEL#
 120679 YIELD#
 62852 OPTIMIZ?
L13     5214 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
      ?)

FILE 'SCISEARCH'
    51760 SERINE
   21683 SER
   151346 RICH
 2077769 HIGH
 1544858 LEVEL#
 395337 YIELD#
 231907 OPTIMIZ?
L14     4738 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
      ?)

```

FILE 'LIFESCI'
 21346 SERINE
 10414 SER
 34806 RICH
 371067 HIGH
 429126 LEVEL#
 54802 YIELD#
 16670 OPTIMIZ?
L15 2904 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

FILE 'BIOTECHDS'
 4795 SERINE
 4602 SER
 4493 RICH
 74156 HIGH
 51021 LEVEL#
 39614 YIELD#
 18716 OPTIMIZ?
L16 601 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

FILE 'BIOSIS'
 68149 SERINE
 22012 SER
 105895 RICH
 1493999 HIGH
 1613796 LEVEL#
 298893 YIELD#
 67102 OPTIMIZ?
L17 5953 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

FILE 'EMBASE'
 56820 SERINE
 18916 SER
 74243 RICH
 1329455 HIGH
 1698265 LEVEL#
 134210 YIELD#
 62502 OPTIMIZ?
L18 4606 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

FILE 'HCAPLUS'
 105605 SERINE
 34615 SER
 275173 RICH
 3744857 HIGH
 2249255 LEVEL#
 1150857 YIELD#
 289102 OPTIMIZ?
L19 9041 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

FILE 'NTIS'
 523 SERINE
 403 SER
 9236 RICH
 328134 HIGH
 228017 LEVEL#
 55396 YIELD#
 59744 OPTIMIZ?
L20 76 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

FILE 'ESBIOBASE'
 26942 SERINE
 12423 SER
 45135 RICH
 510678 HIGH
 573275 LEVEL#
 74897 YIELD#
 30661 OPTIMIZ?
L21 3577 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

FILE 'BIOTECHNO'
 28989 SERINE
 11924 SER
 29372 RICH
 299126 HIGH
 367944 LEVEL#
 41645 YIELD#
 16086 OPTIMIZ?
L22 3241 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

FILE 'WPIDS'
 8297 SERINE
 9747 SER
 33876 RICH
 2046959 HIGH
 610630 LEVEL#
 249912 YIELD#
 41189 OPTIMIZ?
L23 492 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

TOTAL FOR ALL FILES
L24 40443 (SERINE OR SER) (15A) (RICH OR HIGH OR LEVEL# OR YIELD# OR OPTIMIZ
 ?)

=> s l12 and l24

FILE 'MEDLINE'
L25 12 L1 AND L13

FILE 'SCISEARCH'
L26 9 L2 AND L14

FILE 'LIFESCI'
L27 5 L3 AND L15

FILE 'BIOTECHDS'
L28 2 L4 AND L16

FILE 'BIOSIS'
L29 9 L5 AND L17

FILE 'EMBASE'
L30 5 L6 AND L18

FILE 'HCAPLUS'
L31 11 L7 AND L19

FILE 'NTIS'
L32 0 L8 AND L20

FILE 'ESBIOBASE'
L33 4 L9 AND L21

FILE 'BIOTECHNO'
L34 8 L10 AND L22

FILE 'WPIDS'
L35 1 L11 AND L23

TOTAL FOR ALL FILES
L36 66 L12 AND L24

=> s l12 and coexpress?
FILE 'MEDLINE'
13548 COEXPRESS?
L37 2 L1 AND COEXPRESS?

FILE 'SCISEARCH'
13934 COEXPRESS?
L38 2 L2 AND COEXPRESS?

FILE 'LIFESCI'
6199 COEXPRESS?
L39 2 L3 AND COEXPRESS?

FILE 'BIOTECHDS'
660 COEXPRESS?
L40 1 L4 AND COEXPRESS?

FILE 'BIOSIS'
13729 COEXPRESS?
L41 2 L5 AND COEXPRESS?

FILE 'EMBASE'
12791 COEXPRESS?
L42 1 L6 AND COEXPRESS?

FILE 'HCAPLUS'
12757 COEXPRESS?
L43 2 L7 AND COEXPRESS?

FILE 'NTIS'
33 COEXPRESS?
L44 0 L8 AND COEXPRESS?

FILE 'ESBIOBASE'
9809 COEXPRESS?
L45 1 L9 AND COEXPRESS?

FILE 'BIOTECHNO'
7587 COEXPRESS?
L46 1 L10 AND COEXPRESS?

FILE 'WPIDS'
140 COEXPRESS?
L47 0 L11 AND COEXPRESS?

TOTAL FOR ALL FILES
L48 14 L12 AND COEXPRESS?

=> s (amino acid or ser or serine) (15a) (composition# or profil?)
FILE 'MEDLINE'
612125 AMINO
1387745 ACID
459057 AMINO ACID
(AMINO (W) ACID)
21253 SER

89374 SERINE
162979 COMPOSITION#
236020 PROFIL?
L49 13569 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'SCISEARCH'
385550 AMINO
1114189 ACID
204369 AMINO ACID
(AMINO (W) ACID)
21683 SER
51760 SERINE
404372 COMPOSITION#
365251 PROFIL?
L50 9303 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'LIFESCI'
166826 "AMINO"
297172 "ACID"
115086 AMINO ACID
("AMINO" (W) "ACID")
10414 SER
21346 SERINE
97038 COMPOSITION#
52629 PROFIL?
L51 6007 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'BIOTECHDS'
66035 AMINO
135710 ACID
47319 AMINO ACID
(AMINO (W) ACID)
4602 SER
4795 SERINE
40424 COMPOSITION#
10048 PROFIL?
L52 2448 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'BIOSIS'
521001 AMINO
1241250 ACID
303253 AMINO ACID
(AMINO (W) ACID)
22012 SER
68149 SERINE
321074 COMPOSITION#
229122 PROFIL?
L53 21821 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'EMBASE'
421666 "AMINO"
1369373 "ACID"
285316 AMINO ACID
("AMINO" (W) "ACID")
18916 SER
56820 SERINE
146585 COMPOSITION#
199373 PROFIL?
L54 13021 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'HCAPLUS'
1060945 AMINO
4112384 ACID
526592 AMINO ACID
(AMINO (W) ACID)

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34615 SER
105605 SERINE
940605 COMPOSITION#
1384879 COMPN
1938054 COMPOSITION#
    (COMPOSITION# OR COMPN)
422290 PROFIL?
L55      37840 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'NTIS'
6961 AMINO
43883 ACID
2458 AMINO ACID
    (AMINO(W)ACID)
403 SER
523 SERINE
69557 COMPOSITION#
57548 PROFIL?
L56      223 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'ESBIOBASE'
177493 AMINO
336489 ACID
98921 AMINO ACID
    (AMINO(W)ACID)
12423 SER
26942 SERINE
82506 COMPOSITION#
91500 PROFIL?
L57      3439 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'BIOTECHNO'
204625 AMINO
349810 ACID
154660 AMINO ACID
    (AMINO(W)ACID)
11924 SER
28989 SERINE
38895 COMPOSITION#
42958 PROFIL?
L58      6366 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

FILE 'WPIDS'
244392 AMINO
937777 ACID
68710 AMINO ACID
    (AMINO(W)ACID)
9747 SER
8297 SERINE
707136 COMPOSITION#
8956 COMPN
388438 COMPSN
111886 COMPSNS
879081 COMPOSITION#
    (COMPOSITION# OR COMPN OR COMPSN OR COMPSNS)
194379 PROFIL?
L59      4757 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

TOTAL FOR ALL FILES
L60      118794 (AMINO ACID OR SER OR SERINE) (15A) (COMPOSITION# OR PROFIL?)

=> s l12 and l60
FILE 'MEDLINE'
L61      7 L1 AND L49

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FILE 'SCISEARCH'
L62 8 L2 AND L50

FILE 'LIFESCI'
L63 3 L3 AND L51

FILE 'BIOTECHDS'
L64 1 L4 AND L52

FILE 'BIOSIS'
L65 18 L5 AND L53

FILE 'EMBASE'
L66 6 L6 AND L54

FILE 'HCAPLUS'
L67 16 L7 AND L55

FILE 'NTIS'
L68 0 L8 AND L56

FILE 'ESBIOBASE'
L69 3 L9 AND L57

FILE 'BIOTECHNO'
L70 4 L10 AND L58

FILE 'WPIDS'
L71 0 L11 AND L59

TOTAL FOR ALL FILES
L72 66 L12 AND L60

=> s (heterologous or foreign or recombinant) (5a) (protein#) (10a) (produc? or
express? or optimiz?)

FILE 'MEDLINE'
47718 HETEROLOGOUS
59253 FOREIGN
260330 RECOMBINANT
1917452 PROTEIN#
1292191 PRODUC?
987095 EXPRESS?
62852 OPTIMIZ?
L73 9273 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'SCISEARCH'
22183 HETEROLOGOUS
30720 FOREIGN
151971 RECOMBINANT
1506451 PROTEIN#
1820406 PRODUC?
1264806 EXPRESS?
231907 OPTIMIZ?
L74 9497 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'LIFESCI'
14826 HETEROLOGOUS
8447 FOREIGN
66783 RECOMBINANT
557087 PROTEIN#
514871 PRODUC?
391866 EXPRESS?

L75 16670 OPTIMIZ?
 7060 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
 UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'BIOTECHDS'
 10884 HETEROLOGOUS
 6430 FOREIGN
 97090 RECOMBINANT
 151602 PROTEIN#
 222303 PRODUC?
 140581 EXPRESS?
 18716 OPTIMIZ?
L76 28528 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
 UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'BIOSIS'
 29246 HETEROLOGOUS
 27340 FOREIGN
 190479 RECOMBINANT
 1776662 PROTEIN#
 1712468 PRODUC?
 1177112 EXPRESS?
 67102 OPTIMIZ?
L77 11496 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
 UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'EMBASE'
 22283 HETEROLOGOUS
 32257 FOREIGN
 168217 RECOMBINANT
 1528079 PROTEIN#
 1234355 PRODUC?
 900431 EXPRESS?
 62502 OPTIMIZ?
L78 7669 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
 UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'HCAPLUS'
 31811 HETEROLOGOUS
 44935 FOREIGN
 181959 RECOMBINANT
 2145009 PROTEIN#
 4210196 PRODUC?
 925167 PRODN
 4658156 PRODUC?
 (PRODUC? OR PRODN)
 1198161 EXPRESS?
 289102 OPTIMIZ?
L79 21347 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
 UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'NTIS'
 303 HETEROLOGOUS
 384442 FOREIGN
 1594 RECOMBINANT
 18611 PROTEIN#
 370125 PRODUC?
 39074 EXPRESS?
 59744 OPTIMIZ?
L80 139 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
 UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'ESBIOBASE'
 12834 HETEROLOGOUS
 10210 FOREIGN

83652 RECOMBINANT
719877 PROTEIN#
581093 PRODUC?
561398 EXPRESS?
30661 OPTIMIZ?
L81 7785 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'BIOTECHNO'
14199 HETEROLOGOUS
6070 FOREIGN
125134 RECOMBINANT
653195 PROTEIN#
394590 PRODUC?
452182 EXPRESS?
16086 OPTIMIZ?
L82 8130 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
UC? OR EXPRESS? OR OPTIMIZ?)

FILE 'WPIDS'
9977 HETEROLOGOUS
40929 FOREIGN
40069 RECOMBINANT
154449 PROTEIN#
2335750 PRODUC?
124549 EXPRESS?
41189 OPTIMIZ?
L83 4754 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
UC? OR EXPRESS? OR OPTIMIZ?)

TOTAL FOR ALL FILES
L84 115678 (HETEROLOGOUS OR FOREIGN OR RECOMBINANT) (5A) (PROTEIN#) (10A) (PROD
UC? OR EXPRESS? OR OPTIMIZ?)

=> s l12 and l84
FILE 'MEDLINE'
L85 4 L1 AND L73

FILE 'SCISEARCH'
L86 3 L2 AND L74

FILE 'LIFESCI'
L87 5 L3 AND L75

FILE 'BIOTECHDS'
L88 5 L4 AND L76

FILE 'BIOSIS'
L89 2 L5 AND L77

FILE 'EMBASE'
L90 2 L6 AND L78

FILE 'HCAPLUS'
L91 5 L7 AND L79

FILE 'NTIS'
L92 0 L8 AND L80

FILE 'ESBIOBASE'
L93 3 L9 AND L81

FILE 'BIOTECHNO'
L94 2 L10 AND L82

FILE 'WPIDS'
L95 2 L11 AND L83

TOTAL FOR ALL FILES
L96 33 L12 AND L84

=> s 160 and 184
FILE 'MEDLINE'
L97 50 L49 AND L73

FILE 'SCISEARCH'
L98 47 L50 AND L74

FILE 'LIFESCI'
L99 38 L51 AND L75

FILE 'BIOTECHDS'
L100 485 L52 AND L76

FILE 'BIOSIS'
L101 55 L53 AND L77

FILE 'EMBASE'
L102 64 L54 AND L78

FILE 'HCAPLUS'
L103 165 L55 AND L79

FILE 'NTIS'
L104 0 L56 AND L80

FILE 'ESBIOBASE'
L105 42 L57 AND L81

FILE 'BIOTECHNO'
L106 74 L58 AND L82

FILE 'WPIDS'
L107 43 L59 AND L83

TOTAL FOR ALL FILES
L108 1063 L60 AND L84

=> s l108 and coli
FILE 'MEDLINE'
249337 COLI
L109 30 L97 AND COLI

FILE 'SCISEARCH'
228507 COLI
L110 25 L98 AND COLI

FILE 'LIFESCI'
98566 COLI
L111 21 L99 AND COLI

FILE 'BIOTECHDS'
45738 COLI
L112 129 L100 AND COLI

FILE 'BIOSIS'
276667 COLI
L113 29 L101 AND COLI

FILE 'EMBASE'

177298 COLI
L114 34 L102 AND COLI

FILE 'HCAPLUS'
266594 COLI
L115 69 L103 AND COLI

FILE 'NTIS'
2810 COLI
L116 0 L104 AND COLI

FILE 'ESBIOBASE'
68890 COLI
L117 19 L105 AND COLI

FILE 'BIOTECHNO'
94549 COLI
L118 32 L106 AND COLI

FILE 'WPIDS'
18914 COLI
L119 8 L107 AND COLI

TOTAL FOR ALL FILES
L120 396 L108 AND COLI

=> s (l36 or l48 or l72 or l96 or l120)

FILE 'MEDLINE'
L121 51 (L25 OR L37 OR L61 OR L85 OR L109)

FILE 'SCISEARCH'
L122 43 (L26 OR L38 OR L62 OR L86 OR L110)

FILE 'LIFESCI'
L123 32 (L27 OR L39 OR L63 OR L87 OR L111)

FILE 'BIOTECHDS'
L124 133 (L28 OR L40 OR L64 OR L88 OR L112)

FILE 'BIOSIS'
L125 56 (L29 OR L41 OR L65 OR L89 OR L113)

FILE 'EMBASE'
L126 44 (L30 OR L42 OR L66 OR L90 OR L114)

FILE 'HCAPLUS'
L127 98 (L31 OR L43 OR L67 OR L91 OR L115)

FILE 'NTIS'
L128 0 (L32 OR L44 OR L68 OR L92 OR L116)

FILE 'ESBIOBASE'
L129 26 (L33 OR L45 OR L69 OR L93 OR L117)

FILE 'BIOTECHNO'
L130 43 (L34 OR L46 OR L70 OR L94 OR L118)

FILE 'WPIDS'
L131 10 (L35 OR L47 OR L71 OR L95 OR L119)

TOTAL FOR ALL FILES
L132 536 (L36 OR L48 OR L72 OR L96 OR L120)

=> s l132 not 2004-2006/py

FILE 'MEDLINE'

1332548 2004-2006/PY
(20040000-20069999/PY)
L133 44 L121 NOT 2004-2006/PY

FILE 'SCISEARCH'
2454856 2004-2006/PY
(20040000-20069999/PY)
L134 35 L122 NOT 2004-2006/PY

FILE 'LIFESCI'
189530 2004-2006/PY
L135 26 L123 NOT 2004-2006/PY

FILE 'BIOTECHDS'
56959 2004-2006/PY
L136 79 L124 NOT 2004-2006/PY

FILE 'BIOSIS'
1023506 2004-2006/PY
L137 50 L125 NOT 2004-2006/PY

FILE 'EMBASE'
1117159 2004-2006/PY
L138 33 L126 NOT 2004-2006/PY

FILE 'HCAPLUS'
2548350 2004-2006/PY
L139 74 L127 NOT 2004-2006/PY

FILE 'NTIS'
25986 2004-2006/PY
L140 0 L128 NOT 2004-2006/PY

FILE 'ESBIOBASE'
664275 2004-2006/PY
L141 18 L129 NOT 2004-2006/PY

FILE 'BIOTECHNO'
586 2004-2006/PY
L142 43 L130 NOT 2004-2006/PY

FILE 'WPIDS'
2489972 2004-2006/PY
L143 1 L131 NOT 2004-2006/PY

TOTAL FOR ALL FILES
L144 403 L132 NOT 2004-2006/PY

=> dup rem l144
PROCESSING COMPLETED FOR L144
L145 199 DUP REM L144 (204 DUPLICATES REMOVED)

=> d tot

L145 ANSWER 1 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New PRO20080 polypeptides and polynucleotides, useful for treating
immune-related disorders in a mammal, e.g. systemic lupus erythematosus,
rheumatoid arthritis, systemic sclerosis, bullous skin disease, or
allergies;
recombinant protein production and
antagonist and agonist for use in disease gene therapy

AU GREWAL I; GURNEY A L; VALDEZ P A
AN 2003-20959 BIOTECHDS
PI WO 2003055440 10 Jul 2003

- L145 ANSWER 2 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel recombinant crystallized polypeptides from Streptococcus pneumoniae useful as drug target for pathogenic bacteria, has biological activity of NH(3)-dependent NAD(+) synthetase;
plasmid-mediated gene transfer and expression in Escherichia coli for recombinant protein production for recombinant vaccine and disease therapy
- AU EDWARDS A; DHARAMSI A; VEDADI M; ALAM M Z; HOUSTON S; PINDER B; NG I; LAM R; KIMBER M
AN 2003-20545 BIOTECHDS
PI WO 2003051916 26 Jun 2003
- L145 ANSWER 3 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New peptides useful, e.g. in the treatment of or reduction of viral load of hepatitis C virus and associated conditions, e.g. liver fibrosis, necrosis, inflammation or bile duct changes;
vector-mediated gene transfer and expression in host cell for recombinant protein production, drug screening and disease therapy
- AU JOYCE M; WILLIAMS M; HINDSGAUL O; TYRREL D L
AN 2003-22522 BIOTECHDS
PI WO 2003051910 26 Jun 2003
- L145 ANSWER 4 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New GAVE18 polypeptide and nucleic acid molecule encoding the polypeptide, useful for preventing and treating a disease or disorder associated with aberrant expression or activity of GAVE18, e.g. asthma or rheumatoid arthritis;
recombinant protein production and agonist and antagonist for use in disease gene therapy
- AU EISHINGDRELO H; CAI J; BUSCH S J; GASSENHUBER J
AN 2003-17738 BIOTECHDS
PI WO 2003042399 22 May 2003
- L145 ANSWER 5 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New cupiennin peptides exhibiting an antimicrobial, hemolytic or insecticidal activity, useful for preparing a composition for treating bacterial infections and tumors;
vector-mediated gene transfer and expression in host cell for recombinant protein and insecticide production for use in bacterium infection and leukemia gene therapy
- AU SCHALLER J; WALZ A; NENTWIG W; KUHN-NENTWIG L
AN 2003-16957 BIOTECHDS
PI WO 2003035677 1 May 2003
- L145 ANSWER 6 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New polynucleotide designated 205P1B5, for diagnosing and treating prostate cancer, and as probes or primers for the amplification and/or detection of 205P1B5 genes;
recombinant protein production and its encoding gene useful for gene therapy, diagnosis and prognosis
- AU CHALLITA-EID P M; RAITANO A B; FARIS M; HUBERT R S; JAKOBOWITS A
AN 2003-14871 BIOTECHDS
PI WO 2003020954 13 Mar 2003
- L145 ANSWER 7 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New isolated polypeptide based on the neutralizing epitope of the p17 protein of HIV, useful for the diagnosis, prevention and treatment of the human acquired immune deficiency syndrome;
plasmid-mediated gene transfer and expression in Escherichia coli for recombinant glutathione-transferase fusion protein production for use in HIV virus infection therapy

AU CARUSO A; FRANZONE J S
AN 2003-11714 BIOTECHDS
PI WO 2003016337 27 Feb 2003

L145 ANSWER 8 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New UvrA and UvrB polypeptides and polynucleotides encoding the
polypeptides, useful for detecting DNA damage for diagnosing cancer,
increasing the effectiveness of drug treatment or detecting the effect of
environmental genotoxin;
recombinant protein production in
Escherichia coli useful for cancer diagnosis

AU VAN HOUTEN B; SKORVAGA M
AN 2003-11705 BIOTECHDS
PI WO 2003014324 20 Feb 2003

L145 ANSWER 9 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel nitrilase polypeptide, useful for making (R)- or
(S)-ethyl-4-cyano-3-hydroxybutyric acid or (R)- or (S)-mandelic acid or
(S)- or (R)-phenyl lactic acid derivative and for producing
pharmaceutical composition, and food additive;
vector-mediated recombinant protein gene transfer
and expression in host cell for use in pharmaceutical and
food industry and peptidomics

AU MADDEN M; DESANTIS G; CHAPLIN J A; WEINER D P; MILAN A; CHI E; SHORT J M;
BURK M
AN 2003-10320 BIOTECHDS
PI WO 2003000840 3 Jan 2003

L145 ANSWER 10 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New polynucleotide, useful for manipulating plant protein quality,
improving plant growth, yield and crop productivity or grain composition
or producing plants with improved properties;
recombinant protein production via
plasmid expression in host cell for use in transgenic plant
construction

AU EDGERTON M D; CHOMET P S; LACCETTI L B
AN 2004-07435 BIOTECHDS
PI US 2003233670 18 Dec 2003

L145 ANSWER 11 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New nucleic acids encoding PRO polypeptides having sequence identity to
Interleukin-17, useful for diagnosing or treating of immune related
diseases e.g. rheumatoid arthritis, thyroiditis, diabetes mellitus or
allergic rhinitis;

recombinant protein production and
antagonist and agonist for use in disease therapy and gene therapy
AU CHEN J; FILVAROFF E; FONG S; GODDARD A; GODOWSKI P; GRIMALDI J C; GURNEY
A; LI H; HILLAN K; HYMOWITZ S G; TUMAS D; STAROVASNIK M A; VANLOOKEREN M;
VANDLEN R; WATANABE C; WILLIAMS P M; WOOD W I; YANSURA D
AN 2004-05717 BIOTECHDS
PI US 2003203451 30 Oct 2003

L145 ANSWER 12 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New genes from Staphylococcus aureus encode virulence factors designated
repressor of toxin, Rot, and Rot-like protein Rlp and are useful to
detect S. aureus in a sample;
recombinant protein production via
plasmid expression in host cell for in bacterium detection

AU MCNAMARA P J
AN 2004-00325 BIOTECHDS
PI US 2003171563 11 Sep 2003

L145 ANSWER 13 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New estrogen receptor beta variant polypeptide and nucleic acid fragment,
useful in therapeutic modulation of pathophysiologic estrogen signaling

- (e.g. gene delivery or gene silencing), or developing pharmaceutical drug targets;
vector-mediated gene transfer and expression in host cell
for recombinant protein production and
disease therapy or gene therapy
- AU QUINET E M; FAN E
AN 2004-03630 BIOTECHDS
PI US 2003162257 28 Aug 2003
- L145 ANSWER 14 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel vascular endothelial cell growth factor-E polypeptide, useful for
treating cardiovascular or endothelial disorders;
recombinant protein production via
plasmid expression in host cell for use in disease therapy
- AU FERRARA N; KUO S S
AN 2004-01862 BIOTECHDS
PI US 2003113870 19 Jun 2003
- L145 ANSWER 15 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Composition comprising orthogonal aminoacyl-tRNA synthetase that
preferentially aminoacylates orthogonal tRNA with non-natural amino
acids, useful for incorporating non-natural amino acids into polypeptides
in vivo;
recombinant enzyme protein and vector
expression in host cell for use in non-natural amino acid
incorporation and site-selective insertion
- AU SCHULTZ P; WANG L; ANDERSON J C; CHIN J W; LIU D R; MAGLIERI T J; MEGGERS
E L; MEHL R A; PASTRNAK M; SANTORO S W; ZHANG Z
AN 2004-02882 BIOTECHDS
PI US 2003108885 12 Jun 2003
- L145 ANSWER 16 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New purified nucleic acid segment encoding hyaluronic acid synthase,
useful for hyaluronic acid, and for detecting a bacterial cell that is
expressing hyaluronic synthase;
recombinant enzyme protein production
via plasmid expression in host cell useful for bacterium
cell detection
- AU WEIGEL P H; DEANGELIS P L; PAPACONSTANTINOU J
AN 2003-28187 BIOTECHDS
PI US 2003104533 5 Jun 2003
- L145 ANSWER 17 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel purified nucleic acid segment encoding hyaluronic acid synthase,
useful for hyaluronic acid, and for detecting a bacterial cell that is
expressing hyaluronic synthase;
vector-mediated hyaluronic-acid-synthase gene transfer and
expression in host cell for recombinant
protein production and cloning
- AU WEIGEL P H; DEANGELIS P L; PAPACONSTANTINOU J
AN 2003-27658 BIOTECHDS
PI US 2003104415 5 Jun 2003
- L145 ANSWER 18 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel isolated DNA98853 or DNA101848 polypeptide having homology to
members of tumor necrosis factor receptor family useful in assays to
identify proteins or molecules involved in binding interactions;
vector-mediated gene transfer and expression in host cell
for recombinant protein production and
disease therapy
- AU GODDARD A; PAN J; YAN M
AN 2003-28389 BIOTECHDS
PI US 2003092044 15 May 2003
- L145 ANSWER 19 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN

- TI New PRO nucleic acid, useful for preparing a composition for treating an immune related disease in a mammal e.g., rheumatoid arthritis, diabetes mellitus or autoimmune disease;
vector-mediated gene transfer and expression in CHO cell,
yeast or Escherichia coli for recombinant
protein production for use in disease gene therapy
- AU FONG S; GODDARD A; GODOWSKI P J; GRIMALDI J C; GURNEY A L; HILLAN K J;
TUMAS D; WATANABE C K; WOOD W I; ZHANG Z
- AN 2003-22976 BIOTECHDS
- PI US 2003077737 24 Apr 2003
- L145 ANSWER 20 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
- TI Novel isolated PRO polypeptides PRO1265, PRO1308, PRO1475, PRO4405,
PRO5723, PRO7425 or PRO9940, useful for treating an immune-related
disease such as rheumatoid arthritis, osteoarthritis, autoimmune
hemolytic anemia;
recombinant protein production and
antagonist and agonist for use in disease therapy and gene therapy
- AU GODDARD A; GODOWSKI P J; GURNEY A L; HILLAN K J; TUMAS D; WATANABE C K;
WOOD W I
- AN 2003-27366 BIOTECHDS
- PI US 2003059437 27 Mar 2003
- L145 ANSWER 21 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
- TI New isolated nucleic acid encoding a PRO polypeptide, e.g. PRO1031,
PRO1122, PRO10272, useful in molecular biology, chromosome and gene
mapping, in generating antisense RNA and DNA, and in gene therapy;
involving vector-mediated recombinant protein gene
transfer and expression in Chinese hamster ovary,
Escherichia coli or yeast cell for use in diagnosis,
prevention, therapy and gene therapy
- AU CHEN J; FILVAROFF E; FONG S; GODDARD A; GODOWSKI P J; GRIMALDI C; GURNEY
A L; LI H; HILLAN K; TUMAS D; VANLOOKEREN M; VANDLEN R; WATANABE C;
WILLIAMS P M; WOOD W I; YANSURA D G
- AN 2003-14221 BIOTECHDS
- PI US 2003008815 9 Jan 2003
- L145 ANSWER 22 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
- TI Crystalline composition comprising hepatitis C virus (HCV) NS3/NS4A
polypeptide complex, useful for determining three-dimensional structure
of HCV NS3/NS4A complex, and modeling tertiary structure of related
proteins;
protein 3D structure coordinate and antagonist and agonist useful for
drug screening
- AU REICHERT P; PROSISE W W; TAREMI S S; YAO N; WEBER P C
- AN 2003-22442 BIOTECHDS
- PI US 6524589 25 Feb 2003
- L145 ANSWER 23 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
- TI Novel Tumor Necrosis Factor (TNF) delta nucleic acid useful for assaying
genetic variation and aberrations such as defects or to remediate TNF
delta dysfunction, and in gene therapy;
human recombinant protein production and
its encoding gene useful for gene therapy and diagnosis
- AU YU G; NI J; GENTZ R; DILLON P J
- AN 2003-14839 BIOTECHDS
- PI US 6509170 21 Jan 2003
- L145 ANSWER 24 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
- TI Novel protein having D-hydantoinase or D-carbamylase activity, for
manufacturing N-carbamyl-D-amino acids and D-amino acids, in chemical
industry and in food additives;
recombinant enzyme protein production
via plasmid expression in host cell useful for food and
pharmaceutical industry

AN 2003-19878 BIOTECHDS
PI JP 2003024074 28 Jan 2003

L145 ANSWER 25 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Polypeptide ligand of GPR8 comprising N-terminal methionine residue fused
to a polypeptide having N-terminal cysteine residue, useful in the
treatment of cancer and Alzheimer's disease;
recombinant protein production via
plasmid expression in host cell for use in disease therapy
and drug screening

AN 2003-25647 BIOTECHDS
PI JP 2003009867 14 Jan 2003

L145 ANSWER 26 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New PRO polypeptides useful for diagnosing tumor in mammal and for
producing antibodies useful in treatment of neoplastic cell growth;
recombinant protein production and
antibody for use in disease therapy and gene therapy
AU CHEN J; BAKER K P; YUAN J; GURNEY A; GODDARD A; WOOD W I
AN 2004-23473 BIOTECHDS
PI AU 2002330288 17 Apr 2003

L145 ANSWER 27 OF 199 HCPLUS COPYRIGHT 2006 ACS on STN
TI Immunogenic composition containing chimeric HIV polypeptide (p24-gp41 or
p24-gp36), and test-kits for detection of antibodies raised against HIV
SO Russ., No pp. given
CODEN: RUXXE7
IN Sidorovich, I. G.; Nikolaeva, I. A.; Sheval'e, A. F.; Ignat'eva, G. A.;
Korobova, S. V.; Alekseev, T. A.; Petrov, R. V.; Khaitov, R. M.
AN 2003:862724 HCPLUS
DN 140:180118
PATENT NO. KIND DATE APPLICATION NO. DATE

PI RU 2214274 C2 20031020 RU 2001-122896 20010816

L145 ANSWER 28 OF 199 HCPLUS COPYRIGHT 2006 ACS on STN
TI Human stem cell growth factor mutant protein, its preparation and medical
composition
SO Faming Zhuanli Shenqing Gongkai Shuomingshu, 17 pp.
CODEN: CNXXEV
IN Liu, Qingfa; Li, Jing; Hu, Huarong; Hu, Hui
AN 2005:841769 HCPLUS
DN 143:261406
PATENT NO. KIND DATE APPLICATION NO. DATE

PI CN 1445239 A 20031001 CN 2002-111092 20020320

L145 ANSWER 29 OF 199 MEDLINE on STN DUPLICATE 1
TI Engineering *Escherichia coli* for increased productivity of
serine-rich proteins based on proteome profiling
.
SO Applied and environmental microbiology, (2003 Oct) Vol. 69, No. 10, pp.
5772-81.
Journal code: 7605801. ISSN: 0099-2240.
AU Han Mee-Jung; Jeong Ki Jun; Yoo Jong-Shin; Lee Sang Yup
AN 2003497591 MEDLINE

L145 ANSWER 30 OF 199 HCPLUS COPYRIGHT 2006 ACS on STN
TI Quantification of the isomerization of Asp residue in recombinant human
 α A-crystallin by reversed-phase HPLC
SO Journal of Pharmaceutical and Biomedical Analysis (2003), 30(6), 1825-1833
CODEN: JPBADA; ISSN: 0731-7085
AU Sadakane, Yutaka; Yamazaki, Toshiaki; Nakagomi, Kazuya; Akizawa,
Toshifumi; Fujii, Noriko; Tanimura, Takenori; Kaneda, Masaki; Hatanaka,
Yasumaru

AN 2002:951194 HCAPLUS
DN 139:145381

L145 ANSWER 31 OF 199 SCISEARCH COPYRIGHT (c) 2006 The Thomson Corporation
on STN DUPLICATE 2

TI Sulfur assimilation in soybean: Molecular cloning and characterization of
O-acetylserine (thiol) lyase (*cysteine synthase*)
SO CROP SCIENCE, (SEP-OCT 2003) Vol. 43, No. 5, pp. 1819-1827.
ISSN: 0011-183X.

AU Chronis D; Krishnan H B (Reprint)
AN 2003:774749 SCISEARCH

L145 ANSWER 32 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

TI Cloning, expression, and renaturation studies of Reteplase
SO Journal of Microbiology and Biotechnology (2003), 13(6), 989-992
CODEN: JOMBES; ISSN: 1017-7825

AU Zhao, Youchun; Wang, Ge; Kong, Yang; Zhang, Changkai
AN 2004:75941 HCAPLUS
DN 140:247840

L145 ANSWER 33 OF 199 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights
reserved on STN

TI Molecular cloning, expression and characterization of three short chain
 α -neurotoxins from the venom of sea snake - Hydrophiinae *Hydrophis*
cyanocinctus Daudin.

SO Toxicon, (2003) Vol. 42, No. 7, pp. 753-761. .
Refs: 31
ISSN: 0041-0101 CODEN: TOXIA6

AU Peng L.-S.; Zhong X.-F.; Huang Y.-S.; Zhang Y.; Zheng S.-L.; Wei J.-W.; Wu
W.-Y.; Xu A.-L.

AN 2004045475 EMBASE

L145 ANSWER 34 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

TI Cloning and molecular and immunological characterisation of two new food
allergens, Cap a 2 and Lyc e 1, profilins from bell pepper (*Capsicum*
annuum) and tomato (*Lycopersicon esculentum*)

SO International Archives of Allergy and Immunology (2003), 131(4), 245-255
CODEN: IAAIEG; ISSN: 1018-2438

AU Willerroider, M.; Fuchs, H.; Ballmer-Weber, B. K.; Focke, M.; Susani, M.;
Thalhamer, J.; Ferreira, F.; Wuethrich, B.; Scheiner, O.; Breiteneder, H.;
Hoffmann-Sommergruber, K.

AN 2003:623472 HCAPLUS
DN 140:76195

L145 ANSWER 35 OF 199 MEDLINE on STN

TI A novel O-phospho-L-serine sulfhydrylation reaction catalyzed by
O-acetylserine sulfhydrylase from *Aeropyrum pernix* K1.

SO FEBS letters, (2003 Sep 11) Vol. 551, No. 1-3, pp. 133-8.
Journal code: 0155157. ISSN: 0014-5793.

AU Mino Koshiki; Ishikawa Kazuhiko
AN 2003424973 MEDLINE

L145 ANSWER 36 OF 199 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights
reserved on STN DUPLICATE 3

TI Cel6A, a major exoglucanase from the cellulosome of the anaerobic fungi
Piromyces sp. E2 and *Piromyces equi*.

SO Biochimica et Biophysica Acta - Gene Structure and Expression, (9 Jul
2003) Vol. 1628, No. 1, pp. 30-39. .

Refs: 61

ISSN: 0167-4781 CODEN: BBGSD5

AU Harhangi H.R.; Freelove A.C.J.; Ubhayasekera W.; Van Dinther M.;
Steenbakkers P.J.M.; Akhmanova A.; Van Der Drift C.; Jetten M.S.M.;
Mowbray S.L.; Gilbert H.J.; Op Den Camp H.J.M.

AN 2003266875 EMBASE

- L145 ANSWER 37 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New isolated nucleic acid molecule encoding a NS4 polypeptide, useful for
treating a body weight disorder, e.g. obesity, anorexia, cachexia, or
conditions related to obesity, e.g. polycystic ovarian disease,
dermatological disorders;
vector-mediated recombinant protein gene transfer
and expression in Chinese hamster ovary cell culture,
Escherichia coli or yeast cell for use in gene therapy
- AU GODDARD A; PAN J; WOOD W I
AN 2003-10100 BIOTECHDS
PI WO 2002101069 19 Dec 2002
- L145 ANSWER 38 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel immunogenic, mutant cholera holotoxin useful for enhancing immune
response of vertebrate host to antigen, comprises amino sequence of
subunit A of wild-type cholera toxin;
vector-mediated gene transfer and expression in host cell for
recombinant vaccine and immunostimulant
- AU GREEN B A; HOLMES R K; JOBLING M G; ZHU D
AN 2003-09012 BIOTECHDS
PI WO 2002098369 12 Dec 2002
- L145 ANSWER 39 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New TALL-1-binding polypeptide, useful for modulating the activity of
TALL-1 and in treating, preventing or diagnosing a B-cell-mediated
autoimmune diseases, cancers or lymphomas;
vector-mediated recombinant protein gene transfer
and expression in host cell for use in gene therapy
- AU MIN H; HSU H
AN 2003-09315 BIOTECHDS
PI WO 2002092620 21 Nov 2002
- L145 ANSWER 40 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New compound containing acidic and aromatic amino acids, useful as
antiviral therapy in pharmaceutical, veterinary or
agricultural/horticultural applications;
compound production and virus vector expression in host cell for use
in gene therapy
- AU DASGUPTA A; DAS S; BAIDYA N
AN 2003-07297 BIOTECHDS
PI WO 2002083858 24 Oct 2002
- L145 ANSWER 41 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel fusion protein for inducing human immunodeficiency virus-antigen
specific IgG and IgA antibodies, has ectodomain of HIV-1 envelope
glycoprotein gp41 fused to fragment of influenza virus hemagglutinin
protein;
vector-mediated gene transfer and expression in host cell for
recombinant vaccine and HIV virus infection therapy
- AU WEISSENHORN W; WILEY D; MANTIS N; NEUTRA M R; KOZLOWSKI P
AN 2003-07165 BIOTECHDS
PI WO 2002081655 17 Oct 2002
- L145 ANSWER 42 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New PRO842 polypeptides having structural homology to interleukin-8,
useful for treating or diagnosing a mammal with an inflammatory disease
or immune related disease, e.g. rheumatoid arthritis, osteoarthritis or
allergic disease;
vector-mediated gene transfer and expression in host cell
for recombinant protein production, drug
screening and gene therapy
- AU FRENCH D; GRIMALDI J C; HILIAN K J; PISABARRO M T; SCHMIDT K N; SMITH V;
TUMAS D; VANDLEN R L; WATANABE C K; WILLIAMS P M; WOOD W I
AN 2003-05386 BIOTECHDS
PI WO 2002070706 12 Sep 2002

- L145 ANSWER 43 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New product comprising a dendroaspin scaffold and a serine protease inhibitor domain ligated to the dendroaspin scaffold, useful for treating or preventing diseases associated with thrombosis, e.g. myocardial infarction, stroke;
dendroaspin scaffold, serine protease-inhibitor domain and vector expression in host cell use in disease therapy and drug screening
- AU LU X; KAKKAR V V
AN 2003-02235 BIOTECHDS
PI WO 2002063017 15 Aug 2002
- L145 ANSWER 44 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel lipase variant with reduced potential for odor generation for use in detergent compositions, comprises a parent polypeptide having lipase activity and a peptide extension attached to carboxy terminal of the polypeptide;
recombinant protein production via plasmid expression in host cell for surfactant composition
- AU MUNK S; VIND J; BORCH K; PATKAR S A; GLAD S O S; SVENDSEN A
AN 2003-03908 BIOTECHDS
PI WO 2002062973 15 Aug 2002
- L145 ANSWER 45 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New human PRO21074 polypeptide, useful for diagnosing, preventing, treating or monitoring the progression of cartilaginous disorders, e.g. spondyloarthropathies, rheumatoid arthritis or osteoarthritis;
vector-mediated recombinant protein gene transfer and expression in Chinese hamster ovary, Escherichia coli or yeast cell culture for use in disease diagnosis, prevention, therapy and gene therapy
- AU FILVAROFF E; GODDARD A; GRIMALDI J C; WOOD W I
AN 2003-02755 BIOTECHDS
PI WO 2002059308 1 Aug 2002
- L145 ANSWER 46 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel human secreted proteins and polynucleotides, useful for diagnosing and treating tumors, cardiovascular or endothelial disorders, atherosclerosis, cardiac hypertrophy, angiogenic disorders and bone disorders;
vector-mediated recombinant protein gene transfer and expression in host cell for use in gene therapy, recombinant vaccine and nucleic acid vaccine preparation
- AU BASINSKI M B; MICANOVIC R; MILLS B J; SANKHAVARAM P R; SU E W; TSCHANG S R; VARGA G; WANG H
AN 2002-18312 BIOTECHDS
PI WO 2002048361 20 Jun 2002
- L145 ANSWER 47 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel ubiquitin conjugating enzyme polypeptide isolated from activated human T cell, for screening modulators useful for treating cancer, immune disorder, lymphoproliferative disorder, neurodegenerative disorder;
vector-mediated gene transfer, expression in host cell, DNA probe and antibody for recombinant protein production, drug screening and disease therapy
- AU BOWEN M A; WU Y; YANG W; FINGER J N
AN 2002-17873 BIOTECHDS
PI WO 2002036741 10 May 2002
- L145 ANSWER 48 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel chemokine-2 polypeptide and polynucleotide encoding the polypeptide, useful for enhancing immune response to an immunogen in a fish;
vector-mediated recombinant oncogene or cytokine fusion protein gene transfer and expression in host cell

- for disease recombinant vaccine, prognosis, diagnosis and gene therapy
- AU SUNDICK R S; LIU L; DIXON B; FUJIKI K
AN 2002-18256 BIOTECHDS
PI WO 2002036070 10 May 2002
- L145 ANSWER 49 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel proteins and polynucleotides of secreted proteins useful for
treating various diseases e.g. rheumatoid arthritis, cancer, psoriasis,
diabetic retinopathy, arteriosclerosis, ischemia or reperfusion injury;
recombinant protein production and sense
and antisense gene use in disease therapy and gene therapy
- AU SU E W; WANG H
AN 2002-17830 BIOTECHDS
PI WO 2002026801 4 Apr 2002
- L145 ANSWER 50 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Polynucleotide sequences encoding human secretory proteins useful for
gene therapy of e.g. genetic deficiency disorders, cancers, and diseases
caused by intracellular parasites;
recombinant protein gene production via
plasmid expression in host cell, sense, antisense, agonist,
antagonist, transgenic animal, antibody, cell culture, DNA array and
polymerase chain reaction useful in disease gene therapy and drug
screening
- AU STUART J; LINCOLN S E; ALTUS C M; DUFOUR G E; CHALUP M S; HILLMAN J L;
JONES A L; YU J Y; WRIGHT R J; GIETZEN D; LIU T F; YAP P E; DAHL C R;
MOMIYAMA M G; BRADLEY D L; ROHATGI S D; HARRIS B; ROSEBERRY A M; GERSTIN
E H; PERALTA C H; DAVID M H; PANZER S R; FLORES V; DAFFO A; MARWAHA R;
CHEN A J; CHANG S C; AU A P; INMAN R R
AN 2002-12753 BIOTECHDS
PI WO 2002020756 14 Mar 2002
- L145 ANSWER 51 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Identifying targeting peptides useful for treating e.g. diabetes
mellitus, inflammatory diseases, cancer, or autoimmune diseases,
comprises exposing a sample to a phage display library and recovering
phage bound to the sample;
adeno-associated virus vector-mediated recombinant
protein gene transfer and expression in host cell,
Fab and humanized antibody for use in prostate cancer, Hodgkin
disease, diabetes mellitus, inflammatory disease, arthritis,
atherosclerosis, autoimmune disease, bacterium infection, virus
infection, cardiovascular disease and neurodegenerative disease
diagnosis, therapy and gene therapy
- AU ARAP W; PASQUALINI R
AN 2002-13525 BIOTECHDS
PI WO 2002020722 14 Mar 2002
- L145 ANSWER 52 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Monitoring 158P1H4 gene products in biological sample from patient who
has or is suspected of having cancer, useful for treating cancer,
comprises identifying presence of aberrant 158P1H4 gene products in
biological sample;
recombinant protein and monoclonal antibody
production, useful for tumor recombinant vaccine, diagnosis
and prognosis
- AU CHALLITA-EID P M; HUBERT R S; RAITANO A B; AFAR D E H; LEVIN E; FARIS M;
GE W; JAKOBOWITS A
AN 2002-12096 BIOTECHDS
PI WO 2002016598 28 Feb 2002
- L145 ANSWER 53 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel polypeptides and polynucleotides of secreted proteins useful for
treating various diseases such as multiple sclerosis, cancer, autoimmune
diseases, osteoporosis, Alzheimer's disease and Parkinson's disease;

plasmid pQE60-mediated recombinant IgG Fc region chimeric fusion protein gene transfer and expression in Escherichia coli for disease or disorder diagnosis and gene therapy

AU EDMONDS B T; MICANOVIC R; OU W; SU E W; TSCHANG S R; WANG H
AN 2002-12425 BIOTECHDS
PI WO 2002014358 21 Feb 2002

L145 ANSWER 54 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Polymer based intracellular delivery system for protein phosphatases useful for intracellular delivery of protein phosphatases for tumor therapy;
plasmid pET28b vector-mediated recombinant protein gene transfer and expression in Escherichia coli for hydroxypropyl methacrylamide copolymer-PP2C conjugate construction for tumor diagnosis and therapy

AU LAVI S; SATCHE-FAINARO R
AN 2002-13518 BIOTECHDS
PI WO 2002007670 31 Jan 2002

L145 ANSWER 55 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New purified human S-acyl fatty acid synthase thioesterase-like enzyme, useful for identifying modulators of enzyme activity for treating cardiovascular disease, diabetes, obesity and hyperlipidemia;
recombinant protein gene production via plasmid expression in host cell useful in gene therapy

AU XIAO Y
AN 2002-06197 BIOTECHDS
PI WO 2002000855 3 Jan 2002

L145 ANSWER 56 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New isolated IL-17 nucleic acids and polypeptides, useful for diagnosing and treating disorders with aberrant expression or activity of the IL-17 polypeptide, such as degenerative cartilaginous and immune-related disorders;

recombinant protein production and antagonist and agonist for use in disease therapy and gene therapy
AU CHEN J; FILVAROFF E; FONG S; FRENCH D; GODDARD A; GODOWSKI P J; GRIMALDI J C; GURNEY A L; HILLAN K J; HYMOWITZ S G; LI H; PAN J; STAROVASNIK M A; TUMAS D; VAN LOOKEREN M; VANDLEN R; WATANABE C K; WILLIAMS P M; WOOD W I; YANSURA D G
AN 2003-22452 BIOTECHDS
PI US 2002182673 5 Dec 2002

L145 ANSWER 57 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New isolated PAL-18 polypeptide, useful for diagnosing, characterizing, and treating disease and in determining disease susceptibility; vector-mediated recombinant protein gene transfer and expression in host cell for use in mamma cancer diagnosis and therapy

AU KINDERS R J; COREY M J
AN 2003-03761 BIOTECHDS
PI US 2002106765 8 Aug 2002

L145 ANSWER 58 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel isolated Apo-2DcR polypeptide useful for modulating apoptosis in mammalian cells;
plasmid pRK5-mediated recombinant protein gene transfer and expression in CHO cell, yeast, Escherichia coli, 293 cell and HeLa cell and hybridoma cell culture for monoclonal antibody production

AU ASHKENAZI A J; BAKER K P; CHUNTHARAPAI A; GURNEY A; KIM K J; WOOD W I
AN 2003-03758 BIOTECHDS
PI US 2002102706 1 Aug 2002

- L145 ANSWER 59 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel Nodal and Lefty polypeptides useful for diagnosing or treating cell growth and differentiation related disorders in humans, e.g. cancer, autoimmunity, arthritis and immunosuppression;
recombinant protein production and sense and antisense sequence use in gene therapy
- AU EBNER R; SOPPET D R; RUBEN S M
AN 2003-02695 BIOTECHDS
PI US 2002086351 4 Jul 2002
- L145 ANSWER 60 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel antibody specific for a peptide immunogen and comprising at least one streptolysin S epitope, useful as vaccinating agent for eliciting an immune response against streptococcal infections;
vector-mediated recombinant protein gene transfer and expression in host cell for use in recombinant vaccine preparation and Streptococcus sp. infection prevention and therapy
- AU DALE J B
AN 2003-06510 BIOTECHDS
PI US 2002086023 4 Jul 2002
- L145 ANSWER 61 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel apoprotein antigens encoded by Mycoplasma hyopneumoniae for use in vaccines to prevent and treat diseases caused by infection with Mycoplasma hyopneumoniae in animals, especially pigs;
recombinant protein production and sense and antisense sequence use in gene therapy
- AU KING K W; MADURA R A; ROSEY E L
AN 2003-04532 BIOTECHDS
PI EP 1245677 2 Oct 2002
- L145 ANSWER 62 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New protein immobilized at solid phase by binding site, useful for detecting antibodies, comprises multiple antigen/epitope sequences for antibodies, spaced by bridge compositions so that they are exposed for antibody binding;
recombinant protein production and immobilization, vector expression in host cell for antibody detection and virus infection testing
- AU REPKE H; BUDDE E; NICOLAUS S
AN 2003-01093 BIOTECHDS
PI EP 1229044 7 Aug 2002
- L145 ANSWER 63 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New cysD, N, K, E and H genes from coryneform bacteria, useful, when over expressed, for increasing fermentative production of L-amino acids;
vector plasmid pEC-XK99E-mediated recombinant protein gene transfer and expression in Escherichia coli for use in L-amino acid preparation and medicine, pharmaceutical and food industries
- AU FARWICK M; HUTHMACHER K; PFEFFERLE W; SCHISCHKA N; BATHE B
AN 2002-16465 BIOTECHDS
PI DE 10136986 21 Mar 2002
- L145 ANSWER 64 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN
TI Cloning, characterization and biotechnological use of Physcomitrella patens proteins and enzymes involved in the synthesis of amino acids, vitamins, cofactors, nucleotides and nucleosides
SO U.S. Pat. Appl. Publ., 107 pp.
CODEN: USXXCO
IN Lerchl, Jens; Renz, Andreas; Ehrhardt, Thomas; Reindl, Andreas; Cirpus, Petra; Bischoff, Friedrich; Frank, Markus; Freund, Annette; Duwenig, Elke; Schmidt, Ralf-Michael; Reski, Ralf
AN 2002:755097 HCAPLUS
DN 137:275028

| | PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|--------|---|------|----------|-----------------|----------|
| PI | US 2002142422 | A1 | 20021003 | US 2000-734017 | 20001212 |
| L145 | ANSWER 65 OF 199 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN | | | | |
| TI | The sac mutants of Chlamydomonas reinhardtii reveal transcriptional and posttranscriptional control of cysteine biosynthesis | | | | |
| SO | Plant Physiology, (01 DEC 2002), 130/4 (2076-2084), 47 reference(s) | | | | |
| CODEN: | PLPHAY ISSN: 0032-0889 | | | | |
| AU | Ravina C.G.; Chang C.-I.; Tsakraklides G.P.; McDermott J.P.; Vega J.M.; Leustek T.; Gotor C.; Davies J.P. | | | | |
| AN | 2002:36035203 BIOTECHNO | | | | |
| L145 | ANSWER 66 OF 199 MEDLINE on STN | | | | |
| TI | Limits to sulfur accumulation in transgenic lupin seeds expressing a foreign sulfur-rich protein. | | | | |
| SO | Plant physiology, (2002 Mar) Vol. 128, No. 3, pp. 1137-48.
Journal code: 0401224. ISSN: 0032-0889. | | | | |
| AU | Tabe Linda M; Droux Michel | | | | |
| AN | 2002160076 MEDLINE | | | | |
| L145 | ANSWER 67 OF 199 MEDLINE on STN DUPLICATE 4 | | | | |
| TI | Removal of DnaK contamination during fusion protein purifications. | | | | |
| SO | Protein expression and purification, (2002 Aug) Vol. 25, No. 3, pp. 503-7.
Journal code: 9101496. ISSN: 1046-5928. | | | | |
| AU | Rial Daniela V; Ceccarelli Eduardo A | | | | |
| AN | 2002489079 MEDLINE | | | | |
| L145 | ANSWER 68 OF 199 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN | | | | |
| TI | Recombinant human CIS2 (SOCS2) protein: Subcloning, expression, purification, and characterization. | | | | |
| SO | Protein Expression and Purification, (July, 2002) Vol. 25, No. 2, pp. 305-312. print.
CODEN: PEXPEJ. ISSN: 1046-5928. | | | | |
| AU | Biener, Eva; Maurice, Sarah; Sandowski, Yael; Cohen, Yael; Gusakowsky, Eugene E.; Hooghe, Robert; Yoshimura, Akihiko; Livnah, Oded; Gertler, Arieh [Reprint author] | | | | |
| AN | 2002:502157 BIOSIS | | | | |
| L145 | ANSWER 69 OF 199 LIFESCI COPYRIGHT 2006 CSA on STN | | | | |
| TI | Secretory production of recombinant human C-reactive protein in Escherichia coli, capable of binding with phosphorylcholine, and its characterization | | | | |
| SO | Biochemical and Biophysical Research Communications [Biochem. Biophys. Res. Commun.], (20020705) vol. 295, no. 1, pp. 163-166.
ISSN: 0006-291X. | | | | |
| AU | Tanaka, T.; Horio, T.; Matuo, Y. | | | | |
| AN | 2002:103909 LIFESCI | | | | |
| L145 | ANSWER 70 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN | | | | |
| TI | Cloning and characterization of an insecticidal crystal protein gene from Bacillus thuringiensis subspecies kenyaee | | | | |
| SO | Journal of Genetics (2002), 81(1), 5-11
CODEN: JOGNAU; ISSN: 0022-1333 | | | | |
| AU | Misra, Hari S.; Khairnar, Nivedita P.; Mathur, Manjula; Vijayalakshmi, N.; Hire, Ramesh S.; Dongre, T. K.; Mahajan, S. K. | | | | |
| AN | 2002:773523 HCAPLUS | | | | |
| DN | 138:131850 | | | | |
| L145 | ANSWER 71 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN | | | | |
| TI | Isolated PRO1356, PRO617, PRO1030, PRO4302 polypeptides, useful for treating immune disorders such as thyroiditis, diabetes mellitus, allergic disease, asthma, allergic rhinitis, atopic dermatitis; retro virus vector-mediated recombinant protein | | | | |

gene transfer and expression in CHO cell, Escherichia coli and yeast, antagonist, agonist, antisense, DNA primer, DNA probe, monoclonal antibody, humanized antibody, singlechain antibody, expressed sequence tag, database and bioinformatic software for disease diagnosis and gene therapy

AU EATON D L; FONG S; GODDARD A; GODOWSKI P J; GRIMALDI C J; GURNEY A L; WATANABE C K; WOOD W I; ZHANG Z
AN 2002-06124 BIOTECHDS
PI WO 2001092331 6 Dec 2001

L145 ANSWER 72 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel subtilisin protease variants useful in cleaning compositions, e.g. laundry compositions and personal care compositions, has amino acid deletions in defined epitope regions;
phagemid-mediated gene transfer, expression in *Bacillus subtilis* and site-specific mutagenesis for recombinant protein production and cleaner surfactant

AU Rubingh D N; Sikorski E E
AN 2001-06294 BIOTECHDS
PI WO 2001007575 1 Feb 2001

L145 ANSWER 73 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New fertile transgenic maize plant comprises amino acid-elevating amount of expressed recombinant inheritable gene encoding seed storage protein;
plasmid ppHYGI1, plasmid pBII221 and plasmid pZ27Z10-mediated *Escherichia coli* beta-glucuronidase gene transfer and expression in maize transgenic plant for improved herbicide resistance

AU Lundquist R C; Walters D A; Kiriha J A
AN 2002-03070 BIOTECHDS
PI US 2001032344 18 Oct 2001

L145 ANSWER 74 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Composition comprising mutant immunoglobulin (Ig)G molecule having increased half-life relative to IgG, decreasing endogenous serum IgG in a subject, comprises amino acid substitutions in Fc-hinge fragment;
vector-mediated gene transfer, expression in host cell and site-directed mutagenesis for recombinant protein production, vaccine and immunotherapy

AU WARD E S
AN 2002-12089 BIOTECHDS
PI US 6277375 21 Aug 2001

L145 ANSWER 75 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel rhesus receptor having affinity for neuropeptide-Y, pancreatic peptide-P or peptide-YY is useful for identifying compounds for treating disorders and diseases which include cardiovascular conditions and cerebral disorders;
vector-mediated gene transfer, expression in *Escherichia coli* or mammal host cell for recombinant protein production, drug screening and disease therapy or prevention

AU Baez M; Yang P
AN 2001-12439 BIOTECHDS
PI US 6242251 5 Jun 2001

L145 ANSWER 76 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Novel mammalian genes encoding cytokine synthesis inhibitory factor (IL-10) and pharmaceutical compositions containing them for treating diseases associated with cytokine imbalance;
plasmid pCD(SR-a)-mediated interleukin-10 gene transfer and expression in COS-7 cell for recombinant protein production and disease therapy

AU Mosmann T R; Moore K W; Bond M W; Vieira P J M

AN 2001-09374 BIOTECHDS
PI US 6217857 17 Apr 2001

L145 ANSWER 77 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN
TI Human G protein-coupled receptors and their cDNA sequences and tissue expression profiles
SO PCT Int. Appl., 193 pp.
CODEN: PIXXD2

IN Parodi, Luis A.; Lind, Peter; Sejlitz, Torsten

AN 2001:816733 HCAPLUS

DN 135:353834

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------|------|--|-----------------|----------|
| PI WO 2001083553 | A2 | 20011108 | WO 2001-US14050 | 20010501 |
| WO 2001083553 | A3 | 20020822 | | |
| | W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | |
| | RW: | GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG | | |
| AU 2001059322 | A5 | 20011112 | AU 2001-59322 | 20010501 |
| EP 1278844 | A2 | 20030129 | EP 2001-932827 | 20010501 |
| | R: | AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR | | |

L145 ANSWER 78 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

TI Human G protein-coupled receptors and their cDNA sequences and tissue expression profiles

SO PCT Int. Appl., 189 pp.

CODEN: PIXXD2

IN Vogeli, Gabriel

AN 2001:763192 HCAPLUS

DN 135:299596

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------|------|--|-----------------|----------|
| PI WO 2001077330 | A2 | 20011018 | WO 2001-US11330 | 20010406 |
| WO 2001077330 | A3 | 20030206 | | |
| | W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | |
| | RW: | GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG | | |
| AU 2001049922 | A5 | 20011023 | AU 2001-49922 | 20010406 |
| US 2002015998 | A1 | 20020207 | US 2001-828644 | 20010406 |
| EP 1303601 | A2 | 20030423 | EP 2001-923209 | 20010406 |
| | R: | AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR | | |

L145 ANSWER 79 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

TI Recombinant production of active protein in prokaryotes by refolding with reducing agent in arginine containing solution

SO PCT Int. Appl., 57 pp.

CODEN: PIXXD2

IN Yamada, Takao; Tsuji, Isamu; Matsui, Hideki

AN 2001:747994 HCAPLUS

DN 135:300248

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------|---|----------|-----------------|----------|
| PI WO 2001075095 | A1 | 20011011 | WO 2001-JP2712 | 20010330 |
| | W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | |
| | RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG | | | |
| CA 2404567 | AA | 20011011 | CA 2001-2404567 | 20010330 |
| AU 2001044647 | A5 | 20011015 | AU 2001-44647 | 20010330 |
| JP 2001342198 | A2 | 20011211 | JP 2001-99706 | 20010330 |
| EP 1273655 | A1 | 20030108 | EP 2001-917667 | 20010330 |
| | R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR | | | |
| US 2003120042 | A1 | 20030626 | US 2002-240295 | 20020927 |

L145 ANSWER 80 OF 199 LIFESCI COPYRIGHT 2006 CSA on STN
 TI Characterization of the Enzymatic Component of the ADP-Ribosyltransferase Toxin CDTa from Clostridium difficile
 SO Infection and Immunity [Infect. Immun.], (20011000) vol. 69, no. 10, pp. 6004-6011.
 ISSN: 0019-9567.
 AU Guelke, I.; Pfeifer, G.; Liese, J.; Fritz, M.; Hofmann, F.; Aktories, K.; Barth, H.*
 AN 2001:110094 LIFESCI

L145 ANSWER 81 OF 199 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN
 TI Production of β -(pyrazol-1-yl)-L-alanine from L-serine and pyrazol using recombinant Escherichia coli cells expressing serine acetyltransferase and O-acetylsarcosine sulfhydrylase-A
 SO Biotechnology Letters, (2001), 23/24 (2051-2055), 11 reference(s)
 CODEN: BILED3 ISSN: 0141-5492
 AU Mino K.; Yamanoue T.; Ohno K.; Sakiyama T.; Eisaki N.; Matsuyama A.; Nakanishi K.
 AN 2001:34073241 BIOTECHNO

L145 ANSWER 82 OF 199 MEDLINE on STN DUPLICATE 5
 TI cDNA cloning and molecular identification of the major oyster allergen from the Pacific oyster Crassostrea gigas.
 SO Clinical and experimental allergy : journal of the British Society for Allergy and Clinical Immunology, (2001 Aug) Vol. 31, No. 8, pp. 1287-94. Journal code: 8906443. ISSN: 0954-7894.
 AU Leung P S; Chu K H
 AN 2001487002 MEDLINE

L145 ANSWER 83 OF 199 LIFESCI COPYRIGHT 2006 CSA on STN
 TI Rapid Isolation of Monoclonal Antibodies. Monitoring Enzymes in the Phytochelatin Synthesis Pathway
 SO Plant Physiology [Plant Physiol.], (20011100) vol. 127, no. 3, pp. 711-719.
 ISSN: 0032-0889.
 AU Li, Y.; Kandasamy, M.K.; Meagher*, R.B.
 AN 2002:8776 LIFESCI

L145 ANSWER 84 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
 TI Potentiating immune responses to antigens using specified Mycobacterium tuberculosis proteins;
 recombinant protein production for interleukin-12 and interferon-gamma
 AU Skeiky Y
 AN 2000-12157 BIOTECHDS

PI WO 2000039301 6 Jul 2000

L145 ANSWER 85 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New polynucleotides encoding proteins, with e.g. nutritional, chemokine, immune stimulating or suppressing, hematopoiesis regulating, tissue growth, tumor inhibition or antiinflammatory activity; human brain, kidney, blood, bladder, etc. recombinant protein production via vector-mediated gene transfer and expression in mammal host cell for disease therapy
AU Jacobs K; McCoy J M; LaVallie E R; Collins-Racie L A; Evans C; Merberg D; Treacy M; Agostino M J; Steininger II R J; Spaulding V; Wong G G; Clark H F; Fechtel K
AN 2000-06517 BIOTECHDS
PI WO 2000009552 24 Feb 2000

L145 ANSWER 86 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI New polynucleotides encoding human retinoid binding protein especially useful for diagnosing, treating and preventing cancer, inflammation and disorders associated with retinoid metabolism; recombinant protein production via vector plasmid pBluescript-mediated gene transfer and expression in Escherichia coli for diagnosis, therapy and gene therapy
AU Bandman O; Guegler K J; Shah P
AN 2000-07809 BIOTECHDS
PI US 6046027 4 Apr 2000

L145 ANSWER 87 OF 199 WPIDS COPYRIGHT 2006 THE THOMSON CORP on STN
TI An isolated polypeptide conserved in proteobacterial extracellular domains used in the treatment and prevention of bacterial infections.
PI WO 2000061165 A1 20001019 (200062)* EN 83 A61K038-00
RW: AT BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE
W: JP US
IN LUPAS, A N

L145 ANSWER 88 OF 199 MEDLINE on STN DUPLICATE 6
TI Characterization of O-acetyl-L-serine sulfhydrylase purified from an alkaliphilic bacterium.
SO Bioscience, biotechnology, and biochemistry, (2000 Nov) Vol. 64, No. 11, pp. 2352-9.
Journal code: 9205717. ISSN: 0916-8451.
AU Sugihara Y; Yamagata S; Mizuno Y; Ezaki T
AN 2001138968 MEDLINE

L145 ANSWER 89 OF 199 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN DUPLICATE 7
TI Recombinant decorsin: Dynamics of the RGD recognition site.
SO Protein Science, (August, 2000) Vol. 9, No. 8, pp. 1428-1438. print.
ISSN: 0961-8368.
AU Krezel, Andrzej M. [Reprint author]; Ulmer, Jana S.; Wagner, Gerhard; Lazarus, Robert A.
AN 2000:424640 BIOSIS

L145 ANSWER 90 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN
TI Cloning and recombinant expression of insulin receptor ligand-binding domains
SO Shengwu Huaxue Yu Shengwu Wuli Xuebao (2000), 32(6), 627-632
CODEN: SHWPAU; ISSN: 0582-9879
AU Zhang, Hong; Qiao, Zhi-Song; Feng, You-Min
AN 2001:3783 HCAPLUS
DN 135:205742

L145 ANSWER 91 OF 199 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN
TI Expression of a bacterial serine acetyltransferase in transgenic potato plants leads to increased levels of cysteine

- and glutathione
SO Plant Journal, (2000), 22/4 (335-343), 54 reference(s)
CODEN: PLJUED ISSN: 0960-7412
AU Harms K.; Von Ballmoos P.; Brunold C.; Hofgen R.; Hesse H.
AN 2000:30400021 BIOTECHNO
- L145 ANSWER 92 OF 199 MEDLINE on STN DUPLICATE 8
TI Molecular cloning and functional characterization of cDNAs encoding cysteine synthase and serine acetyltransferase that may be responsible for high cellular cysteine content in Allium tuberosum.
SO Gene, (2000 Oct 31) Vol. 257, No. 2, pp. 269-77.
Journal code: 7706761. ISSN: 0378-1119.
AU Urano Y; Manabe T; Noji M; Saito K
AN 20001061875 MEDLINE
- L145 ANSWER 93 OF 199 SCISEARCH COPYRIGHT (c) 2006 The Thomson Corporation on STN
TI Regulation of sulfate transport and synthesis of sulfur-containing amino acids
SO CURRENT OPINION IN PLANT BIOLOGY, (JUN 2000) Vol. 3, No. 3, pp. 188-195.
ISSN: 1369-5266.
AU Saito K (Reprint)
AN 2000:377187 SCISEARCH
- L145 ANSWER 94 OF 199 HCPLUS COPYRIGHT 2006 ACS on STN
TI Extracellular Expression, Purification, and Characterization of a Winter Flounder Antifreeze Polypeptide from Escherichia coli
SO Protein Expression and Purification (2000), 18(2), 175-181
CODEN: PEXPEJ; ISSN: 1046-5928
AU Tong, Li; Lin, Qingsong; Wong, W. K. Raymond; Ali, Asma; Lim, Daniel; Sung, Wing L.; Hew, Choy L.; Yang, Daniel S. C.
AN 2000:127507 HCPLUS
DN 132:344551
- L145 ANSWER 95 OF 199 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN DUPLICATE 9
TI Characterization of Api g 1.0201, a new member of the Api g 1 family of celery allergens.
SO International Archives of Allergy and Immunology, (2000) Vol. 122, No. 2, pp. 115-123. .
Refs: 49
ISSN: 1018-2438 CODEN: IAAIEG
AU Hoffmann-Sommergruber K.; Ferris R.; Pec M.; Radauer C.; O'Riordain G.; Laimer da Camara Machado M.; Scheiner O.; Breiteneder H.
AN 2000238357 EMBASE
- L145 ANSWER 96 OF 199 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN DUPLICATE 10
TI Trichomonas vaginalis: Characterization, expression, and phylogenetic analysis of a carbamate kinase gene sequence.
SO Experimental Parasitology, (2000) Vol. 95, No. 1, pp. 54-62. .
Refs: 42
ISSN: 0014-4894 CODEN: EXPAAA
AU Minotto L.; Edwards M.R.; Bagnara A.S.
AN 2000225429 EMBASE
- L145 ANSWER 97 OF 199 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN
TI O-acetylserine sulfhydrylase from Methanosaarcina thermophila
SO Journal of Bacteriology, (2000), 182/1 (45-50), 49 reference(s)
CODEN: JOBAAY ISSN: 0021-9193
AU Borup B.; Ferry J.G.
AN 2000:30004556 BIOTECHNO
- L145 ANSWER 98 OF 199 MEDLINE on STN DUPLICATE 11

- TI Sequence polymorphism of the group 1 allergen of Bermuda grass pollen.
 SO Clinical and experimental allergy : journal of the British Society for
 Allergy and Clinical Immunology, (1999 Apr) Vol. 29, No. 4, pp. 488-96.
 Journal code: 8906443. ISSN: 0954-7894.
 AU Chang Z N; Peng H J; Lee W C; Chen T S; Chua K Y; Tsai L C; Chi C W; Han S
 H
 AN 1999220064 MEDLINE
- L145 ANSWER 99 OF 199 SCISEARCH COPYRIGHT (c) 2006 The Thomson Corporation
 on STN DUPLICATE 12
 TI Physicochemical studies of hepatitis A virus recombinant proteins:
 interaction with monolayers as membrane models
 SO MATERIALS SCIENCE & ENGINEERING C-BIOMIMETIC AND SUPRAMOLECULAR SYSTEMS,
 (1 DEC 1999) Vol. 8-9, Sp. iss. SI, pp. 481-485.
 ISSN: 0928-4931.
 AU Carmona M A; Alsina M A; Pinto R M; Sanchez G; Guix S; Pujol M (Reprint)
 AN 2000:40699 SCISEARCH
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L145 ANSWER 29 OF 199 MEDLINE on STN DUPLICATE 1
AB Variations in proteome profiles of *Escherichia coli* in response to the overproduction of human leptin, a serine-

rich (11.6% of total amino acids) protein, were examined by two-dimensional gel electrophoresis. The levels of heat shock proteins increased, while those of protein elongation factors, 30S ribosomal protein, and some enzymes involved in amino acid biosynthesis decreased, after leptin overproduction. Most notably, the levels of enzymes involved in the biosynthesis of serine family amino acids significantly decreased. Based on this information, we designed a strategy to enhance the leptin productivity by manipulating the *cysK* gene, encoding cysteine synthase A. By coexpression of the *cysK* gene, we were able to increase the cell growth rate by approximately twofold. Also, the specific leptin productivity could be increased by fourfold. In addition, we found that *cysK* coexpression can improve the production of another serine-rich protein, interleukin-12 beta chain, suggesting that this strategy may be useful for the production of other serine-rich proteins as well. The approach taken in this study should be useful in designing a strategy for improving recombinant protein production.

L145 ANSWER 64 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

AB Isolated nucleic acid mols., designated metabolic pathway protein (MP) nucleic acid mols., which encode novel MP proteins from *Phycomitrella patens* are described. The cDNA sequences and the encoded amino acid sequences of a number of MP enzymes and proteins are disclosed. The invention also provides antisense nucleic acid mols., recombinant expression vectors containing MP protein nucleic acid mols., and host cells into which the expression vectors have been introduced. The invention still further provides isolated MP proteins, mutated MP proteins, fusion proteins, antigenic peptides and methods for the improvement of production of a desired compound from transformed cells, organisms or plants based on genetic engineering of MP protein genes in these organisms.

L145 ANSWER 68 OF 199 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

AB The 1x myc-tagged cDNA encoding for human CIS2 protein was subcloned into a pET-29a+ vector in order to express and produce a recombinant S-peptide tagged and 1x myc-tagged protein in *Escherichia coli* BL21(DE3). The constitutively expressed protein was isolated from inclusion bodies by a simple solubilization-renaturation procedure and purified by anion-exchange chromatography on Q-Sepharose. The recombinant form was found to be pure and monomeric as judged by both SDS-PAGE and gel-filtration chromatography and its biological activity was proven by its ability to bind to the tyrosine-phosphorylated cytosolic fragment of human growth hormone receptor fused to glutathione-S-transferase. Recombinant CIS2 was compared by biochemical, immunological, and molecular methods to the CIS2 protein expressed in eukaryotic cells. This report describes the first substantial production of biologically active recombinant human CIS2.

L145 ANSWER 69 OF 199 LIFESCI COPYRIGHT 2006 CSA on STN

AB Recombinant human CRP (rhCRP) was secreted into culture supernatant of *Escherichia coli* by co-expressing kil gene that has a function to secrete colicin E1 outside the cell. Highly purified 5 g rhCRP was produced from 180 L culture supernatant by affinity chromatography. The purified rhCRP was indistinguishable from the native one with respect to Ca super(2+) -dependent binding ability to phosphorylcholine, electrophoretic behavior, N-terminal amino acid analysis, and immunochemical properties. The molecular weight of rhCRP monomer was determined to be 23059.7 Da by TOF/MS analysis. These results indicate that rhCRP has the same protein structure as native one and that rhCRP has the potential as a reference material and/or calibrator of high-sensitivity CRP assay to predict the risk of cardiovascular disease.
[copy] 2002 Elsevier Science (USA)

L145 ANSWER 70 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

AB A sporulating culture of *Bacillus thuringiensis* subsp. *kenyae* strain HD549 is toxic to larvae of lepidopteran insect species such as *Spodoptera litura*, *Helicoverpa armigera* and *Phthorimaea operculella*, and a dipteran insect, *Culex fatigans*. A 1.9-kb DNA fragment, PCR-amplified from HD549 using cryll-gene-specific primers, was cloned and expressed in *E. coli*. The recombinant protein produced 92% mortality in first-instar larvae of *Spodoptera litura* and 86% inhibition of adult emergence in *Phthorimaea operculella*, but showed very low toxicity against *Helicoverpa armigera*, and lower mortality against third-instar larvae of dipteran insects *Culex fatigans*, *Anopheles stephensi* and *Aedes aegypti*. The sequence of the cloned crystal protein gene showed almost complete homol. with a mosquitocidal toxin gene from *Bacillus thuringiensis* var. *kurstaki*, with only five mutations scattered in different regions. Amino acid alignment with different insecticidal crystal proteins using the MUTALIN program suggested presence of the conserved block 3 region in the sequence of this protein. A mutation in codon 409 of this gene that changes a highly conserved phenylalanine residue to serine lies in this block.

L145 ANSWER 81 OF 199 BIOTECHNO COPYRIGHT 2006 Elsevier Science B.V. on STN

AB β -(Pyrazol-1-yl)-L-alanine (β -PA) was produced from L-serine and pyrazol using recombinant *Escherichia coli* cells expressing serine acetyltransferase and O-acetylsine sulfhydrylase-A. The amount of β -PA increased with increasing L-serine concentrations up to 600 mM at 50 mM pyrazol while 100 mM pyrazol gave the highest β -PA production with 50 mM L-serine. Under the optimized conditions, β -PA accumulated in the broth at approximately 140 mM with a conversion of 90% with respect to the added amount of pyrazol.

L145 ANSWER 94 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

AB HPLC6 is the major component of liver-type antifreeze polypeptides (AFPs) from the winter flounder, *Pleuronectes americanus*. To facilitate mutagenesis studies of this protein, a gene encoding the 37-amino acid mature polypeptide was chemical synthesized and cloned into the Tac cassette immediately after the bacterial *ompA* leader sequence for direct excretion of the AFP into the culture medium. *Escherichia coli* transformant with the construct *placiQpar8AF* was cultured in M9 medium. The recombinant AFP (rAFP) was detected by a competitive ELISA. After IPTG induction, a biol. active rAFP was expressed. The majority of the rAFP was excreted into the culture medium with only trace amts. trapped in the periplasmic space and cytoplasm. After 18 h of induction, the accumulated rAFP in the culture medium amounted to about 16 mg/L. The excreted AFP was purified from the culture medium by a single-step reverse-phase HPLC. Mass spectrometric and amino acid composition analyses confirmed the identity of the purified product. The rAFP, which lacked amidation at the C-terminal, was about 70% active when compared to the amidated wild-type protein, thus confirming the importance of C-terminal cap structure in protein stability and function.
(c) 2000 Academic Press.

L145 ANSWER 101 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

AB The consensus repeat sequence found in the dragline silk from the spider, *Nephila clavipes*, was redesigned to incorporate a redox trigger flanking the β -sheet-forming polyalanine sequences. The methionine redox trigger, in the oxidized state, was incorporated to prevent the formation of the β -sheets; in the reduced state it would not result in steric limitations to β -sheet formation. A synthetic gene incorporating the trigger was constructed, cloned, and expressed in *Escherichia coli*. The purified protein, .apprx.25 kDa, contained the expected amino acid composition and migration behavior on SDS-PAGE. The recombinant protein was analyzed by x-ray diffraction, TEM, electron diffraction, and CD in both oxidized and reduced states. Based on the results, the incorporation of a redox trigger appears to be a powerful exptl. strategy to explore the self-assembly of fibrous proteins

such as silks.

L145 ANSWER 105 OF 199 HCPLUS COPYRIGHT 2006 ACS on STN

AB The combination of 2-D PAGE, computer image anal., and several protein identification techniques allowed the Escherichia coli SWISS-2DPAGE database to be established. This is part of the ExPASy mol. biol. server accessible through the WWW at the URL address <http://www.expasy.ch/ch2d/ch2d-top.html>. The authors report recent progress in the development of the E. coli SWISS-2DPAGE database. Proteins were separated with immobilized pH gradients in the 1st dimension and Na dodecyl sulfate-polyacrylamide gel electrophoresis in the 2nd dimension. To increase the resolution of the separation and thus the number of identified proteins, a variety of wide and narrow range immobilized pH gradients were used in the 1st dimension. Micropreparative gels were electroblotted onto polyvinylidene difluoride membranes and spots were visualized by amido black staining. Protein identification techniques such as amino acid composition anal., gel comparison and microsequencing were used, as well as a recently described Edman sequence tag approach. Some of the above identification techniques were coupled with database searching tools. Currently 231 polypeptides are identified on the E. coli SWISS-2DPAGE map: 64 were identified by N-terminal microsequencing, 39 by amino acid composition, and 82 by sequence tag. Of 153 proteins putatively identified by gel comparison, 65 were confirmed. Many proteins were identified using more than 1 technique. Faster progress in the E. coli proteome project will now be possible with advances in biochem. methodol. and with the completion of the entire E. coli genome.

L145 ANSWER 106 OF 199 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN DUPLICATE 16

AB The psoriasis-associated fatty acid binding protein (PA-FABP, also known as FABP5) is a novel keratinocyte protein that is highly up-regulated in psoriatic plaques (P. Madsen, H.H. Rasmussen, H. Leffers, B. Honore and J.E. Celis, J. Invest. Dermatol. 1992, 99, 299-305). Here we have expressed PA-FABP in Escherichia coli as a fusion protein containing an NH₂-terminal hexa-His tag followed by a factor Xa cleavage site. The recombinant protein was expressed at a level of about 30% of the soluble proteins and was purified to homogeneity using a simple two-step protocol consisting of affinity chromatography on Ni²⁺-nitrilotriacetic acid agarose followed by gel filtration. The recombinant protein was then digested with factor Xa and characterized by two-dimensional gel electrophoresis. The ability of PA-FABP to bind saturated fatty acids ranging from 6 to 16 carbons was determined directly by dialysis and compared to human serum albumin (HSA). The results showed that PA-FABP binds multiple molecules of the fatty acids hexanoate (C(6:)), octanoate (C(8:0)), decanoate (C(10:0)) and laurate (C(12:0)), all with a K₁ of about 104 M⁻¹, and myristate (C(14:0)) with a K₁ of 4.4 X 10⁵ M⁻¹. Palmitate (C(16:)) also bound strongly with multiple molecules. Due to the very low solubility of palmitate its affinity to PA-FABP was measured relatively to HSA and found to be 8.1 times lower. At ligand/protein ratios below 1, all fatty acids bound to PA-FABP with about one to three orders of magnitude lower affinity than to HSA. The difference in the fatty acid binding properties of the two proteins may reflect differences in their three-dimensional structures, which in the case of PA-FABP consists mainly of β-sheets while HSA contains predominantly α-helices.

L145 ANSWER 107 OF 199 LIFESCI COPYRIGHT 2006 CSA on STN

AB Coexpression of di- alpha -globin and beta -globin in Escherichia coli in the presence of exogenous heme yielded high levels of soluble, functional recombinant human hemoglobin (rHb1.1). High-level expression of rHb1.1 provides a good model for measuring mistranslation in heterologous proteins. rHb1.1 does not contain isoleucine; therefore, any isoleucine present could be attributed to mistranslation, most likely mistranslation of one or more of

the 200 codons that differ from an isoleucine codon by 1 bp. Sensitive amino acid analysis of highly purified rHb1.1 typically revealed less than or equal to 0.2 mol of isoleucine per mol of hemoglobin. This corresponds to a translation error rate of less than or equal to 0.001, which is not different from typical translation error rates found for *E. coli* proteins. Two different expression systems that resulted in accumulation of globin proteins to levels equivalent to similar to 20% of the level of *E. coli* soluble proteins also resulted in equivalent translational fidelity.

L145 ANSWER 110 OF 199 SCISEARCH COPYRIGHT (c) 2006 The Thomson Corporation
on STN DUPLICATE 19

AB The amplified expression of a recombinant protein is known to lend to an intracellular depletion of specific amino acid pools which in turn may affect the production of the desired protein. In order to counteract and overcome such a situation during the fermentation of the recombinant *Escherichia coli* (PMSG27) containing the glucose isomerase (GI) gene from *Streptomyces* sp. NCIM 2730, the effect of addition of different amino acids on the specific activity of GI was studied. The amino acid composition of GI from *Streptomyces* sp. NCIM 2730 reveals predominantly aspartic acid glutamic acid, and glycine; therefore, in the present paper; the effect of coordinated addition of the assorted combinations of these three amino acids on the synthesis of recombinant GI was studied. The results were analyzed using a 2(3) factorial design. The following conclusions were derived from the analysis of two-factor interactions of the three amino acids: (i) The interaction between the aspartic and glutamic acid is independent of aspartic acid concentration but is affected by the increasing concentrations of glutamic acid, (ii) The effect of aspartic acid concentration is more than that of glycine, and (iii) During the interaction of glutamic acid and glycine, the effect of glutamic acid is more prominent than that of glycine. The three-factor interaction analyses reveal that the effect of the three amino acids is in the order aspartic acid > glutamic acid > glycine. (C) 1998 Elsevier Science Inc.

L145 ANSWER 114 OF 199 LIFESCI COPYRIGHT 2006 CSA on STN

AB Isolation of the recombinant protein from a genetically engineered *Escherichia coli* 1854 producer for further chemical enzymatic transformation into human insulin through proinsulin was studied. Under optimal conditions, the recombinant protein formation was more than 35% of the total cell proteins. Structures of the polypeptides obtained and purified chromatographically were confirmed by amino acid analysis. Human proinsulin was derived from the recombinant protein isolated.

L145 ANSWER 119 OF 199 MEDLINE on STN DUPLICATE 24

AB A partial cDNA clone, from the 3' end of the dragline silk gene was isolated from *Nephila clavipes* major ampullate glands. This clone contains a 1.7-kb insert, consisting of a repetitive coding region of 1.4-kb and a 0.3-kb nonrepetitive coding region; 1.5-kb of the 1.7-kb fragment was cloned into *Escherichia coli* and a 43-kDa recombinant silk protein was expressed. Characterization of the purified protein by Western blot, amino acid composition analysis, and matrix-assisted laser desorption ionization/time-of-flight mass spectrometry confirms it to be spider dragline silk.

L145 ANSWER 127 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

AB Previous research has shown that overexpression of recombinant proteins places a metabolic burden on the host cell. Anal. shows that recombinant proteins expressed in *Escherichia coli* at less than 10% of total protein have an average amino acid composition which is rich in amino acids from the aromatic and serine biosynthetic families. A neg. correlation is observed

between expression level and aromatic amino acid and cysteine content relative to the host. We hypothesize that when an amino acid is present in a recombinant protein at levels significantly higher than present in host cellular protein, the amino acid becomes a limiting factor in expression level. Results will be presented which suggest that 1) the amino acid composition of a recombinant protein can affect its expression level, and 2) the metabolic burden imposed by amino acid compn . can be alleviated by supplementing the cell with required precursors, leading to significant increases in recombinant protein expression.

L145 ANSWER 128 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

AB Gilthead seabream (*S. aurata*) insulin-like growth factor-I (gsIGF-I) cDNA coding for the mature protein was cloned in a pGEM-3Z vector, and then transferred into prokaryotic expression vector pET-11a and expressed in *E. coli* BL21(DE3) cells upon induction with iso-Pr thiogalactoside. The expressed protein contained within the inclusion-body pellet was solubilized in 4.5M urea, refolded for 24 h at pH 11.3 in the presence of catalytic amts. of cysteine, and purified to >98% purity, as a monomeric methionyl-gsIGF-I. Amino acid composition and N-terminal sequence confirmed the identity to be the predicted protein. Binding assays of the ¹²⁵I-gsIGF-I to gilthead seabream or carp (*Cyprinus carpio*) sera resulted in high specific binding, indicating the existence of ≥ 1 IGF-binding proteins. In binding expts. to crude gilthead seabream brain homogenate, using human (h) IGF-I, as a ligand, the resp. IC₅₀ value of hIGF-I was .apprx.4-fold lower than that of gsIGF-I. Recombinant gsIGF-I exhibited mitogenic activity in a mouse mammary gland-derived MME-L1 cell line which was .apprx.200-fold lower than that of hIGF-I. Binding expts. to intact MME-L1 cells suggests that this difference most likely results from a correspondingly lower affinity for IGF-I receptor in these cells. In contrast, the activities of gsIGF-I and hIGF-I measured by ³⁵S uptake by gill arches from the goldfish (*Carassius auratus*) were identical, indicating that the recombinant gsIGF-I is biol. active.

L145 ANSWER 132 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN

AB Previous research has shown that overexpression of recombinant proteins places a metabolic burden on the host cell. Analysis shows that recombinant proteins expressed in *Escherichia coli* at less than 10% of total protein have an average amino acid composition which is rich in amino acids from the aromatic and serine biosynthetic families. A negative correlation was observed between expression level and aromatic amino acid and cysteine content relative to the host. It was hypothesized that when an amino acid is present in a recombinant protein at levels significantly higher than present in host cellular protein, the amino acid becomes a limiting factor in expression level. Results were presented which suggest that the amino acid composition of a recombinant protein can affect its expression level and the metabolic burden imposed by amino acid composition can be alleviated by supplementing the cell with required precursors, leading to significant increases in recombinant protein production . (0 ref)

L145 ANSWER 136 OF 199 SCISEARCH COPYRIGHT (c) 2006 The Thomson Corporation on STN

AB Fed-batch culture with controlled L-amino acid composition was performed to improve production of a recombinant gene product in *Bacillus brevis*. The maximum recombinant protein (alpha-amylase) level and specific activity increased from 5.14 kU/mL and 0.77 kU/mg dry cell in conventional fed-batch culture to 12.01 kU/mL and 2.64 kU/mg dry cell, respectively, when L-amino acid concentration was controlled at 5 mM using an asparagine (Asn)- and isoleucine

(Ile)-enriched nitrogen source. The L-amino acid concentration in the culture was monitored by an automatic biotech analyzer and controlled at 2-20 mM using a mixture of polypeptone and yeast extract. Although L-amino acid concentrations were controlled at low levels, the alpha-amylase activity increased only 1.3 times compared to an uncontrolled batch culture; accumulation of ammonium ion was not reduced. When L-amino acid was controlled at the high level, more cell mass and less recombinant gene product were produced than in those with low control level. To overcome ammonium ion inhibition, the specific amino acids Asn and lie were substituted to improve the production of gene product. Addition of these amino acids to a flask culture led to an improvement in the enzyme production level and specific activity to 2.9 and 5.1 times, respectively, as high as that without them. Both the control of amino acids at low concentrations and the enrichment of Asn and lie were effective for the improvement of recombinant protein production from recombinant *B. brevis* cells. (C) 1996 John Wiley & Sons, Inc.

L145 ANSWER 137 OF 199 HCAPLUS COPYRIGHT 2006 ACS on STN

AB Heterologous proteins may be expressed in bacteria, where protein engineering techniques can be used to alter the amino acid sequence. As the amino acid compn . of the mol. shifts, it is sometimes possible to identify proteins with augmented properties or to identify domains crucial for biol. function. We have developed expression systems that make it possible to produce recombinant proteins at high yield from *E. coli* where partitioning of the product into a cellular compartment can complement a purification strategy. We have now developed an expression system for the production of small peptides by recombinant means at high yield, including when the peptide is a component of a larger fusion protein. Data will be presented on the recombinant production and purification of antifungal peptides from bacteria. In addition, we linked antifungal peptides to a protein carrier to initiate peptide engineering studies with these recombinant fusion proteins. As with recombinant proteins, peptide fusions can be used to characterize and evolve the functional peptide unit.

L145 ANSWER 149 OF 199 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN

AB The ice nucleation-active protein of *Erwinia ananas* IN-10 (inaA protein) was over-expressed as inclusion bodies in *Escherichia coli* YA21 harboring plasmid pINA6S13. The inaA protein was purified from inclusion bodies by repeated solubilization with Triton X-100 to obtain a protein preparation free from sugar and lipid. The yield was 15.3 mg of inaA protein from 60 mg of bacterial cells on a dry weight basis. The N-terminal amino acid sequence of the purified inaA protein was Met-Lys-Glu-Asp-Lys-Val-Leu-Ile-Leu, which agreed exactly with that predicted from the inaA gene. The amino acid composition corresponded approximately with that predicted from the sequence of the inaA gene. The small deviation from the predicted value may have been a result of the very high mol.weight, 130,000, of the inaA protein. The purified protein showed ice nucleation activity, indicating that the inaA protein per se was able to act as an ice nucleus. The study establishes a simplified and enlarged system for the production of pure inaA protein. The protein could be used for the mild freeze-concentration of foods and other materials sensitive to deep-freezing. (20 ref)

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FILE 'HCAPLUS'

L164 3 L152 AND L7

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L167 1 L155 AND L10

FILE 'WPIDS'

L168 1 L156 AND L11

TOTAL FOR ALL FILES

L169 19 L157 AND L12

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PROCESSING COMPLETED FOR L169

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L170 ANSWER 1 OF 5 MEDLINE on STN DUPLICATE 1
TI Differential gene expression for investigation of Escherichia coli biofilm
inhibition by plant extract ursolic acid.
SO Applied and environmental microbiology, (2005 Jul) Vol. 71, No. 7, pp.
4022-34.
Journal code: 7605801. ISSN: 0099-2240.
AU Ren Dacheng; Zuo Rongjun; Gonzalez Barrios Andres F; Bedzyk Laura A;
Eldridge Gary R; Pasmore Mark E; Wood Thomas K
AN 2005348397 MEDLINE

L170 ANSWER 2 OF 5 HCAPLUS COPYRIGHT 2006 ACS on STN DUPLICATE 2
TI Effect of treatment with 4,5-dihydroxy-2-cyclopenten-1-one (DHCP) on gene
expression and **quorum-sensing** in bacteria
SO PCT Int. Appl., 37 pp.
CODEN: PIXXD2

IN Phadtare, Sangita; Kato, Ikunoshin; Inouye, Masayori
AN 2003:757467 HCAPLUS
DN 139:271016

| | PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|----|---------------|--|----------|-----------------|----------|
| PI | ----- | ----- | ----- | ----- | ----- |
| PI | WO 2003077844 | A2 | 20030925 | WO 2003-US7081 | 20030307 |
| | WO 2003077844 | A3 | 20040325 | | |
| | W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
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LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH,
PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ,
UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW | | | | |
| RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY,
KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES,
FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR,
BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | | |
| EP 1482793 | A2 | 20041208 | EP 2003-713993 | 20030307 |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK | | | | |
| JP 2005519616 | T2 | 20050707 | JP 2003-575898 | 20030307 |
| US 2006035317 | A1 | 20060216 | US 2005-506778 | 20050601 |

L170 ANSWER 3 OF 5 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
 TI Growth-phase dependency of autoinducer-2 synthesis in *Helicobacter pylori*.
 SO Abstracts of the General Meeting of the American Society for Microbiology,
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 Meeting Info.: 103rd American Society for Microbiology General Meeting.
 Washington, DC, USA. May 18-22, 2003. American Society for Microbiology.
 ISSN: 1060-2011 (ISSN print).
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 Forsyth, M. H. [Reprint Author]
 AN 2003:519056 BIOSIS

L170 ANSWER 4 OF 5 MEDLINE on STN DUPLICATE 3
 TI The luxS gene is involved in cell-cell signalling for toxin production in *Clostridium perfringens*.
 SO Molecular microbiology, (2002 Apr) Vol. 44, No. 1, pp. 171-9.
 Journal code: 8712028. ISSN: 0950-382X.
 AU Ohtani Kaori; Hayashi Hideo; Shimizu Tohru
 AN 2002230093 MEDLINE

L170 ANSWER 5 OF 5 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
 TI Characterization of quorum sensing pathways in *E. coli*.
 SO Abstracts of the General Meeting of the American Society for Microbiology,
 (1999) Vol. 99, pp. 363. print.
 Meeting Info.: 99th General Meeting of the American Society for
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 ISSN: 1060-2011.
 AU Rather, P. N. [Reprint author]; Baca-Delancey, R. R. [Reprint author];
 Ding, X. [Reprint author]
 AN 1999:311307 BIOSIS

=> d ab 3-5

L170 ANSWER 3 OF 5 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
 AB Background: *Helicobacter pylori* is one of a diverse array of bacterial species with a density-dependent signal system. We previously demonstrated that *H. pylori* produces autoinducer-2 (AI-2) as a quorum sensing molecule and that luxS is involved in its production. LuxS is the third gene in a putative three-gene operon (*cysK*, *metB*, *luxS*). *cysK* and *metB* encode enzymes critical to amino acid synthesis. The mechanism of synthesis and detection of AI-2 by *H. pylori* has not fully been elucidated, and the relationship between production of AI-2 and growth phase has received little attention. We hypothesize that *CysK*, *MetB*, and *LuxS* are part of the same metabolic pathway. Additionally, this study aims to examine the kinetics of AI-2 production in *H. pylori*. Methods: In order to characterize the production of AI-2 in *H. pylori* during other phases of growth, we isolated conditioned media (CM) from cultures of *H. pylori* J99 and 26695 at several

points during growth. The CM generated was assayed for AI-2 levels utilizing a *Vibrio harveyi* bioassay. We have generated a 26695 derivative, L26-3, that is a luxS merodiploid, containing an ectopic copy of luxS in ureA. We have also generated a *H. pylori* strain 26695 derivative (L26-4) with the genotype *cysK*::CAT ure::luxS/ *aphA3*. 26695, L26-3 and L26-4 were grown in broth to mid-log phase, and CM was generated. This CM was assayed for AI-2 using a *V. harveyi* bioassay. Results: While previous results have demonstrated that *H. pylori* strain 60190 has maximal AI-2 production in mid-log phase and that AI-2 production is greatly reduced in stationary phase cultures, we find that *H. pylori* strains 26695 and J99 putatively continue to produce AI-2 through stationary phase. Preliminary analysis of L26-3 revealed increased levels of AI-2 production. Analysis of L26-4 CM suggests that *cysK* may not be essential for AI-2 synthesis. Conclusions: The variation in AI-2 production through the phases of culture growth suggests that the production of AI-2 varies by *H. pylori* strain. Further analysis of the pathway leading to AI-2 production may serve to clarify the significance of this molecule in regulating gene expression in *H. pylori*.

L170 ANSWER 4 OF 5 MEDLINE on STN DUPLICATE 3
AB A Gram-positive anaerobic pathogen, *Clostridium perfringens*, causes clostridial myonecrosis or gas gangrene in humans by producing numerous extracellular toxins and enzymes that act in concert to degrade host tissues. *C. perfringens* possesses a homologue of the luxS gene that is reported to be responsible for the production of autoinducer 2 (AI-2), which participates in quorum sensing in bacteria. The luxS mutant was constructed using *C. perfringens* strain 13, and the role of the luxS gene in toxin production was examined. The cell-free culture supernatant from wild-type strain 13 greatly stimulated the luminescence of *Vibrio harveyi* BB170, whereas that from the luxS mutant caused no significant stimulation, indicating that the luxS gene is necessary for AI-2 production in *C. perfringens*. The luxS mutant showed a reduced level of production of alpha-, kappa- and theta-toxins. In the luxS mutant, the transcription of the theta-toxin gene (*pfoA*) was lower at mid-exponential growth phase, whereas alpha- and kappa-toxin gene transcription was not significantly affected. The production of toxins in the luxS mutant was stimulated by the addition of the culture supernatant from the wild-type cells, possibly because of the presence of AI-2. Moreover, the expression of the *pfoA* gene in the luxS mutant was apparently activated when the mutant cells were cultured in the presence of culture supernatants from the wild-type *C. perfringens*, *Escherichia coli* DH5alpha carrying the luxS gene of *C. perfringens*. A deletion analysis of the luxS operon showed that the luxS gene alone is responsible for cell-cell signalling, and that the metB or *cysK* genes located upstream of luxS are not involved in regulating toxin production. Our results indicate that cell-cell signalling by AI-2 plays an important role in the regulation of toxin production in *C. perfringens*.

L170 ANSWER 5 OF 5 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN

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L171      0 L146 AND L49

FILE 'SCISEARCH'
L172      0 L147 AND L50

FILE 'LIFESCI'
L173      0 L148 AND L51

FILE 'BIOTECHDS'
L174      0 L149 AND L52

FILE 'BIOSIS'
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L175 O L150 AND L53

FILE 'EMBASE'

L176 1 L151 AND L54

FILE 'HCAPLUS'

L177 0 L152 AND L55

FILE 'NTIS'

L178 0 L153 AND L56

FILE 'ESBIOBASE'

L179 0 L154 AND L57

FILE 'BIOTECHNO'

L180 0 L155 AND L58

FILE 'WPIDS'

L181 0 L156 AND L59

TOTAL FOR ALL FILES

L182 1 L157 AND L60

=> d

L182 ANSWER 1 OF 1 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN

TI Acyl-homoserine lactone acylase from Ralstonia strain XJ12B represents a novel and potent class of quorum-quenching enzymes.

SO Molecular Microbiology, (2003) Vol. 47, No. 3, pp. 849-860. .
Refs: 58

ISSN: 0950-382X CODEN: MOMIEE

AU Lin Y.-H.; Xu J.-L.; Hu J.; Wang L.-H.; Leong Ong S.; Renton Leadbetter J.; Zhang L.-H.

AN 2003041693 EMBASE

=> d ab

L182 ANSWER 1 OF 1 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN

AB N-acylhomoserine lactones (AHLs) are used as signal molecules by many quorum-sensing Proteobacteria. Diverse plant and animal pathogens use AHLs to regulate infection and virulence functions. These signals are subject to biological inactivation by AHL-lactonases and AHL-acylases. Previously, little was known about the molecular details underlying the latter mechanism. An AHL signal-inactivating bacterium, identified as a Ralstonia sp., was isolated from a mixed-species biofilm. The signal inactivation encoding gene from this organism, which we callaiID, was cloned and successfully expressed in Escherichia coli and inactivated three AHLs tested. The predicted 794-amino-acid polypeptide was most similar to the aculeacin A acylase (AAC) from *Actinoplanes utahensis* and also shared significant similarities with cephalosporin acylases and other N-terminal (Ntn) hydrolases. However, the most similar homologues of AiiD are deduced proteins of undemonstrated function from available Ralstonia, Deinococcus and Pseudomonas genomes. LC-MS analyses demonstrated that AiiD hydrolyses the AHL amide, releasing homoserine lactone and the corresponding fatty acid. Expression of AiiD in *Pseudomonas aeruginosa* PAO1 quenched quorum sensing by this bacterium, decreasing its ability to swarm, produce elastase and pyocyanin and to paralyse nematodes. Thus, AHL-acylases have fundamental implications and hold biotechnological promise in quenching quorum sensing.

=> s l157 and l84
FILE 'MEDLINE'
L183 8 L146 AND L73

FILE 'SCISEARCH'
L184 10 L147 AND L74

FILE 'LIFESCI'
L185 9 L148 AND L75

FILE 'BIOTECHDS'
L186 7 L149 AND L76

FILE 'BIOSIS'
L187 8 L150 AND L77

FILE 'EMBASE'
L188 9 L151 AND L78

FILE 'HCAPLUS'
L189 10 L152 AND L79

FILE 'NTIS'
L190 0 L153 AND L80

FILE 'ESBIOBASE'
L191 12 L154 AND L81

FILE 'BIOTECHNO'
L192 8 L155 AND L82

FILE 'WPIDS'
L193 1 L156 AND L83

TOTAL FOR ALL FILES
L194 82 L157 AND L84

=> s l194 not 2004-2006/py
FILE 'MEDLINE'
1332548 2004-2006/PY
(20040000-20069999/PY)
L195 6 L183 NOT 2004-2006/PY

FILE 'SCISEARCH'
2454856 2004-2006/PY
(20040000-20069999/PY)
L196 6 L184 NOT 2004-2006/PY

FILE 'LIFESCI'
189530 2004-2006/PY
L197 8 L185 NOT 2004-2006/PY

FILE 'BIOTECHDS'
56959 2004-2006/PY
L198 4 L186 NOT 2004-2006/PY

FILE 'BIOSIS'
1023506 2004-2006/PY
L199 6 L187 NOT 2004-2006/PY

FILE 'EMBASE'
1117159 2004-2006/PY
L200 7 L188 NOT 2004-2006/PY

FILE 'HCAPLUS'

L201 2548350 2004-2006/PY
6 L189 NOT 2004-2006/PY

FILE 'NTIS'
L202 25986 2004-2006/PY
0 L190 NOT 2004-2006/PY

FILE 'ESBIOBASE'
L203 664275 2004-2006/PY
9 L191 NOT 2004-2006/PY

FILE 'BIOTECHNO'
L204 586 2004-2006/PY
8 L192 NOT 2004-2006/PY

FILE 'WPIDS'
L205 2489972 2004-2006/PY
0 L193 NOT 2004-2006/PY

TOTAL FOR ALL FILES
L206 60 L194 NOT 2004-2006/PY

=> dup rem 1206
PROCESSING COMPLETED FOR L206
L207 14 DUP REM L206 (46 DUPLICATES REMOVED)

=> d tot

L207 ANSWER 1 OF 14 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Method of regulating genes in microorganisms, useful for e.g. increasing
production of recombinant proteins, comprises
treating gene-containing microorganism with 4,5-dihydroxy-2-cyclopenten-1-
one;
gene expression regulation and DNA array useful for
recombinant protein promotion
AU PHADTARE S; KATO I; INOUYE M
AN 2003-25814 BIOTECHDS
PI WO 2003077844 25 Sep 2003

L207 ANSWER 2 OF 14 LIFESCI COPYRIGHT 2006 CSA on STN DUPLICATE 1
TI Characterization of RAP, a quorum sensing activator of
Staphylococcus aureus
SO FEMS Microbiology Letters [FEMS Microbiol. Lett.], (20030627) vol. 223,
no. 2, pp. 167-175.
ISSN: 0378-1097.
AU Korem, M.; Sheoran, A.S.; Gov, Y.; Tzipori, S.; Borovok, I.; Balaban, N.*
AN 2003:84905 LIFESCI

L207 ANSWER 3 OF 14 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
TI Crystal of LuxS protein which is involved in production of autoinducer-2
for identifying modulators useful for treating e.g. infection disease,
stomach cancer, stomach ulcer and other intestinal complications;
vector-mediated gene transfer, expression in host cell and
computer bioinformatic software for recombinant
protein production and drug screening
AU LEWIS H A
AN 2002-18835 BIOTECHDS
PI WO 2002038595 16 May 2002

L207 ANSWER 4 OF 14 MEDLINE on STN DUPLICATE 2
TI Genes encoding the N-acyl homoserine lactone-degrading enzyme are
widespread in many subspecies of Bacillus thuringiensis.
SO Applied and environmental microbiology, (2002 Aug) Vol. 68, No. 8, pp.
3919-24.
Journal code: 7605801. ISSN: 0099-2240.

- AU Lee Sang Jun; Park Sun-Yang; Lee Jung-Ju; Yum Do-Young; Koo Bon-Tag; Lee Jung-Kee
AN 2002398079 MEDLINE
- L207 ANSWER 5 OF 14 LIFESCI COPYRIGHT 2006 CSA on STN DUPLICATE 3
TI LuxS-dependent quorum sensing in *Porphyromonas gingivalis* modulates protease and haemagglutinin activities but is not essential for virulence
SO Microbiology, (20020300) vol. 148, no. 3, pp. 763-772.
ISSN: 1350-0872.
AU Burgess, N.A.; Kirke, D.F.; Williams, P.; Winzer, K.; Hardie, K.R.; Meyers, N.L.; Aduse-Opoku, J.; Curtis, M.A.; Camara, M.
AN 2002:77596 LIFESCI
- L207 ANSWER 6 OF 14 MEDLINE on STN DUPLICATE 4
TI Effect of glycine on the cell yield and growth rate of *Escherichia coli*: evidence for cell-density-dependent glycine degradation as determined by (13)C NMR spectroscopy.
SO Journal of biotechnology, (2002 Jan 18) Vol. 92, No. 3, pp. 237-49.
Journal code: 8411927. ISSN: 0168-1656.
AU Han Ling; Doverskog Magnus; Enfors Sven-Olof; Haggstrom Lena
AN 2001639835 MEDLINE
- L207 ANSWER 7 OF 14 EMBASE COPYRIGHT (c) 2006 Elsevier B.V. All rights reserved on STN
TI Bacterial autoinduction: Looking outside the cell for new metabolic engineering targets.
SO Microbial Cell Factories, (20 Dec 2002) Vol. 1, pp. 9p. .
Refs: 91
ISSN: 1475-2859 CODEN: MCFICT
AU DeLisa M.P.; Bentley W.E.
AN 2004253557 EMBASE
- L207 ANSWER 8 OF 14 HCAPLUS COPYRIGHT 2006 ACS on STN
TI Bacterial autoinduction: looking outside the cell for new metabolic engineering targets
SO Microbial Cell Factories (2002), 1, No pp. given
CODEN: MCFICT; ISSN: 1475-2859
URL: <http://www.microbialcellfactories.com/content/pdf/1475-2859-1-5.pdf>
AU DeLisa, Matthew P.; Bentley, William E.
AN 2004:815256 HCAPLUS
DN 141:422094
- L207 ANSWER 9 OF 14 MEDLINE on STN DUPLICATE 5
TI Mapping stress-induced changes in autoinducer AI-2 production in chemostat-cultivated *Escherichia coli* K-12.
SO Journal of bacteriology, (2001 May) Vol. 183, No. 9, pp. 2918-28.
Journal code: 2985120R. ISSN: 0021-9193.
AU DeLisa M P; Valdes J J; Bentley W E
AN 2001270821 MEDLINE
- L207 ANSWER 10 OF 14 MEDLINE on STN DUPLICATE 6
TI Quorum signaling via AI-2 communicates the "Metabolic Burden" associated with heterologous protein production in *Escherichia coli*.
SO Biotechnology and bioengineering, (2001 Nov 20) Vol. 75, No. 4, pp. 439-50.
Journal code: 7502021. ISSN: 0006-3592.
AU DeLisa M P; Valdes J J; Bentley W E
AN 2001565037 MEDLINE
- L207 ANSWER 11 OF 14 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
TI Effect of glycine on the cell yield and growth rate of *Escherichia coli*: Evidence for cell-density-dependent glycine degradation as determined by

13C NMR spectroscopy.
SO Journal of Biotechnology, (18 January, 2001) Vol. 92, No. 3, pp. 237-249.
print.
CODEN: JBITD4. ISSN: 0168-1656.
AU Han, Ling; Doverskog, Magnus; Enfors, Sven-Olof; Haggstrom, Lena [Reprint
author]
AN 2002:147032 BIOSIS

L207 ANSWER 12 OF 14 Elsevier BIOBASE COPYRIGHT 2006 Elsevier Science B.V.
on STN
AN 2001034881 ESBIOBASE
TI Co-overexpression of RspAB improves recombinant protein
production in Escherichia coli
AU Weikert C.; Canonaco F.; Sauer U.; Bailey J.E.
CS U. Sauer, Institute of Biotechnology, ETH Zurich, CH-8093 Zurich,
Switzerland.
E-mail: sauer@biotech.biol.ethz.ch
SO Metabolic Engineering, (2000), 2/4 (293-299), 34 reference(s)
CODEN: MEENFM ISSN: 1096-7176
DT Journal; Article
CY United States
LA English
SL English

L207 ANSWER 13 OF 14 MEDLINE on STN DUPLICATE 7
TI Characterization of the SarA virulence gene regulator of Staphylococcus
aureus.
SO Molecular microbiology, (1999 Jul) Vol. 33, No. 2, pp. 307-16.
Journal code: 8712028. ISSN: 0950-382X.
AU Rechtin T M; Gillaspy A F; Schumacher M A; Brennan R G; Smeltzer M S;
Hurlburt B K
AN 1999340210 MEDLINE

L207 ANSWER 14 OF 14 MEDLINE on STN DUPLICATE 8
TI Characterization of a novel RNA regulator of Erwinia carotovora ssp.
carotovora that controls production of extracellular enzymes and secondary
metabolites.
SO Molecular microbiology, (1998 Jul) Vol. 29, No. 1, pp. 219-34.
Journal code: 8712028. ISSN: 0950-382X.
AU Liu Y; Cui Y; Mukherjee A; Chatterjee A K
AN 1998367138 MEDLINE

=> d ab tot

L207 ANSWER 1 OF 14 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN
AB DERWENT ABSTRACT:
NOVELTY - Method for regulating a gene (I) in a microorganism comprises
treating gene-containing microorganism with 4,5-dihydroxy-2-cyclopenten-1-
one (DHCP). (I) encodes a ribosomal protein or a protein of known or
unknown function or is involved in the response to stress, membrane
synthesis or function, or general metabolism.
DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for: (1)
a method (M1) of screening for physiologically active compounds (A) by
determining the expression level of genes that are modulated by DHCP; (2)
(A) identified by (M1); (3) a method (M2) for increasing production of a
recombinant polypeptide in a microorganism by culturing it in presence of
DHCP; (4) a method (M3) for inhibiting an activity of an interspecies
quorum-sensing inducer, using DHCP; and (5) a
composition, containing DHCP, for: (a) regulating quorum
sensing; (b) regulating expression of specific genes;
(c) promoting secretion of recombinant proteins; or
(d) maintaining homeostasis.
ACTIVITY - None given.
MECHANISM OF ACTION - Gene Expression Regulator. Escherichia coli

JM83 was incubated in presence of 250 microM 4,5-dihydroxy-2-cyclopenten-1-one (DHCP), then RNA isolated and used to probe DNA arrays that represent all open-reading frames of E. coli K-12 W3110. Expression levels were compared with those in untreated cells. Most genes encoding ribosomal L proteins were downregulated (treated:control ratio 0.23-0.49); also (1) 40 genes encoding cell membrane-related proteins were affected, mostly downregulated but some, e.g. creD (unknown function) and tehA (tellurite resistance) were upregulated; (2) the rpoS gene, and genes regulated by the RpoS (a global stress-response regulator) protein were upregulated; (3) 44 genes involved in general metabolism were affected, particularly those implicated in Cys biosynthesis were upregulated and (4) 54 other genes, some of unknown function, were also modified, especially genes involved in resistance to tellurite and oxidative stress were upregulated. DHCP also eliminates the effect of the **quorum-sensing** inducer AI-2.

USE - The method uses DHCP as a regulator of a wide range of bacterial genes to: (1) manufacture a composition for regulating a gene in a microorganism; (2) inhibit activity of interspecies **quorum-sensing** inducers (**quorum-sensing** is associated with virulence, motility and outer membrane function); (3) promote secretion of recombinant proteins; and (4) maintain homeostasis (all claimed). (37 pages)

L207 ANSWER 2 OF 14 LIFESCI COPYRIGHT 2006 CSA on STN DUPLICATE 1

AB **S**taphylococcus aureus are Gram-positive bacteria and cause diverse serious diseases in humans and animals through the production of toxins. The production of toxins is regulated by **quorum sensing** mechanisms, where proteins such as RNAlII activating protein (RAP) are secreted by the bacteria and induce virulence. Antibodies to RAP have been shown to protect mice from infection, but the molecular structure of RAP was not known and hindered vaccine development. To characterize RAP, recombinant protein was made and tested for its ability to induce genes important for pathogenesis (agr). In addition, monoclonal antibodies were produced to identify its cellular localization. Results shown here indicate that RAP is a 277-aa protein that is an ortholog of the ribosomal protein L2. Like the native molecule, **recombinant RAP** activates the **production** of RNAlII (encoded by agr). Using RAP specific monoclonal antibodies we demonstrate that RAP is continuously secreted and while RAP is expressed also in other bacteria (like *Staphylococcus epidermidis*, *Staphylococcus xylosus* and *Escherichia coli*), it is secreted to the culture medium only by *S. aureus*. Our results show that the ribosomal protein L2 has an extraribosomal function and that when secreted RAP acts as an autoinducer of virulence to regulate *S. aureus* pathogenesis.

L207 ANSWER 3 OF 14 BIOTECHDS COPYRIGHT 2006 THE THOMSON CORP. on STN

AB DERWENT ABSTRACT:

NOVELTY - A crystal (I) comprising LuxS protein (which is involved in the production of autoinducer-2 (AI-2), an intercellular signaling molecule employed in the **quorum sensing** pathway of various bacteria) or a functional LuxS protein subunit in crystalline form, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following: (1) a crystal (II) comprising a homolog of LuxS protein having a root mean square deviation of the alpha-carbon atoms of less than 2.0 Angstrom; (2) making (M1) (II) by mixing a volume of a solution comprising the LuxS protein with a volume of a reservoir solution comprising a precipitant and incubating the mixture obtained over the reservoir solution in a closed container, under conditions suitable for crystallization until the crystal forms; (3) determining (M2) the three-dimensional structure of a LuxS protein crystal, by providing (I) or (II) and analyzing the crystal by X-ray diffraction; (4) a machine-readable medium embedded with: (a) information that corresponds to a three-dimensional structural representation of (I) or (II); (b) molecular structure coordinates as shown in the specification or at least

50% of the coordinates; or (c) molecular structure coordinates of a protein molecule comprising LuxS protein binding pocket comprising at least three amino acids from Glu60, Arg68, Ile81 and Asp80, Ala64, His61, Tyr91, Ser9, Phe10 and Leu7, His14, Arg23, Asp40, Arg42, Met84, Cys86 and Thr88 having the structure coordinate as shown in the specification or by the structure coordinates of a binding pocket homolog where the root mean square deviation of the backbone atoms of the amino acid residues of the binding pocket and the binding pocket homolog is less than 2.0 Angstrom; (5) producing (M3) a mutant of LuxS protein having altered property related to LuxS protein by constructing a three-dimensional structure of LuxS protein having structure coordinates of (I)/(II); using modeling methods to identify in the three-dimensional structure at least one structural portion of the LuxS protein molecule, where an alteration in the structural portion is predicted to result in the altered property; providing a nucleic acid molecule having a modified sequence that encodes a deletion, insertion, or substitution of one or more amino acids at a position corresponding to the structural portion; and expressing the nucleic acid molecule to produce the mutant; (6) identifying (M4) a candidate binding compound capable of binding to the active site (or accessory binding site) of LuxS protein, by introducing into a computer program information derived from structural coordinates defining an active site (or accessory binding site) conformation of a LuxS protein molecule based upon three-dimensional structure determination comprising an active site (or accessory binding site) formed by at least the interaction of amino acids Glu, Arg, Ile and Asp where the program utilizes or displays their three-dimensional structure; generating a three-dimensional representation of the active site (or accessory binding site) cavity of the LuxS protein in the computer program; superimposing a model of the binding test compound on the model of the active site (or accessory binding site) of the LuxS protein; and assessing whether the test compound model fits spatially into the active site (or accessory binding site) of the LuxS protein; (7) selecting (M5) at least one compound that potentially binds to LuxS protein, by: (a) constructing a three-dimensional structure of LuxS protein and selecting at least one compound which potentially binds LuxS protein; (b) constructing a three-dimensional structure of a protein molecule comprising a LuxS protein binding pocket and computationally screening several compounds using the structure constructed; and (c) computationally screening a three-dimensional structural representation of a molecule comprising a LuxS protein binding pocket an identifying those that bind; (8) designing (M6) a compound that modulates LuxS protein activity by providing a computer modeling program with a set of structure coordinates, or a three-dimensional conformation derived from them, for a molecule that comprises a binding pocket having the structural coordinates of the binding pocket of LuxS protein, or a binding pocket homolog; computationally building a chemical entity represented by set of structure coordinates and determining whether the chemical entity is a modulator expected to bind to or interfere with the molecule; (9) a compound (C1) identified, designed or made by M4, M5 and M6; (10) a pharmaceutical composition comprising C1 or its salt and a carrier; (11) obtaining structural information about a molecule or a molecular complex of unknown structure by crystallizing the molecule or molecular complex; generating an x-ray diffraction pattern from the crystallized molecule or molecular complex and using a molecular replacement method to interpret the structure of the molecule, where the molecular replacement method uses the structure coordinates as given in the specification, or its subset, or the structure coordinates of the binding pocket; and (12) homology modeling a LuxS protein homolog by: (a) aligning the amino acid sequence of LuxS protein homolog with an amino acid sequence of LuxS protein; (b) incorporating the sequence of homolog into a model of the structure of LuxS protein; (c) subjecting the preliminary model to energy minimization to yield an energy minimized model; and (d) remodeling regions of the energy minimized model where stereochemistry restraint are violated to yield a final model of the homolog.

BIOTECHNOLOGY - Preferred Crystal: (I) is preferably diffraction

quality, is an apo-crystal, a native crystal, and/or is a heavy-atom derivative crystal, where LuxS is Helicobacter pylori, Haemophilus influenzae or Deinococcus radiodurans LuxS, or a mutant which is selenomethionine, selenocysteine mutant, conservative mutant, truncated or extended mutant. (I) is characterized by a set of structure coordinate that is substantially similar to the set of structure coordinates as given in the specification. (II) is produced by mixing a volume of a solution comprising the LuxS protein with a volume of a reservoir solution comprising a precipitant and incubating the mixture obtained over the reservoir solution in a closed container, under conditions suitable for crystallization until the crystal forms. Protein co-ordinate data is given in the patent specification. Preferred Method: In M3, the altered activity of LuxS protein is preferably altered binding activity or immunogenicity, where an epitope is altered. In M4, the structural coordinates correspond to the liganded or unliganded LuxS protein, and the binding compound is a LuxS inhibitor. M5 further comprises screening a library of compounds. The binding pocket comprises at least three amino acids from Glu60, Arg68, Ile81 and Asp80, Ala64, His61, Tyr91, Ser9, Phe10 and Leu7, His14, Arg23, Asp40, Arg42, Met84, Cys86 and Thr88 having the structure coordinate as shown in the specification or a molecule comprising a binding pocket homolog where the root mean square deviation of the backbone atoms of the amino acid residues of the binding pocket and the binding pocket homolog is less than 2.0 Angstrom. The method comprises determining whether the compound potentially binds to the molecule by performing a fitting operation between the compound and a binding pocket of the molecule or molecular complex, and computationally analyzing the results of the fitting operation to quantify the association between, or the interference with, the compound and the binding pocket.

ACTIVITY - Antibacterial; Cytostatic; Antiulcer. No supporting biological data is given.

MECHANISM OF ACTION - LuxS protein modulator (claimed). No supporting biological data is given.

USE - C1 is useful for modulating LuxS protein activity (claimed), useful for treating e.g. infection disease, stomach cancer, stomach ulcer and other intestinal complications.

ADMINISTRATION - C1 is administered through oral, buccal, sublingual, rectal, transdermal, vaginal, transmucosal, nasal or intestinal administration, parenteral delivery, including intramuscular, subcutaneous, intramedullar injections, as well as intrathecal, direct intraventricular, intravenous, intraperitoneal, intranasal or intraocular injections. Dosage of C1 is for 0.01-1000 (preferably 10-30) mg/day.

EXAMPLE - An open-reading frame for LuxS was amplified from *Helicobacter pylori* (Hp-ATCC43504D) genomic DNA by the polymerase chain reaction (PCR) using the following primers: Forward primer GGATTCACATATGAAAATGAATGTAGAGAGTTTC, Reverse Primer: GTTCGGATCCAACCCCCACTTCAGACC. The PCR product (456 bp expected) was digested with NdeI and BamHI, electrophoresed on a 1% agarose gel in TBE buffer and the appropriate size band was excised from the gel and eluted using a standard gel extraction kit. The eluted DNA was ligated overnight with T4 DNA ligase at 16 degreesC into pSB3, previously digested with NdeI and BamHI. The vector pSB3 was a modified version of pET26b where the following sequence had been inserted into the BamHI siteL GGATCCCACCACCACCACCTGAGATCC. The resulting sequence of the gene after being ligated into the vector, from the Shine-Dalgarno sequence through the stop site and the original BamHI, site was as follows: AAGGAGGAGATATACATATG(open reading frame (ORF))GGATCCCACCACCACCACTGAGATCC. The LuxS expressed using this vector had 8 amino acids to the C-terminal end (Gly-Ser-His-His-His-His-His). Plasmids containing ligated inserts were transformed into chemically competent *Escherichia coli* such as Top 10 cells. Colonies were then screened for inserts in the correct orientation and miniprepped. The miniprep DNA was transformed into BL21 (DE3) Active Motif cells and plated onto petri dishes containing Luria-Bertani medium (LB) agar with 30 mug/ml of kanamycin. Isolated, single colonies were grown to mid-log phase and stored at -80

degrees Centigrade in LB containing 15% glycerol. LuxS containing selenomethionine was overexpressed in Escherichia coli and the cultures were allowed to ferment overnight and the LuxS was purified. For crystals of Helicobacter pylori from which the molecular structure coordinates of were obtained, it had been found that a hanging drop containing 1 microlitre of LuxS polypeptide 5 mg/mL in 10 mM HEPES pH 7.5, 150 mM NaCl, 1 mM betaME, 10 mM methionine, and 1 microlitre reservoir solution 32% (w/v) PEG1000, 200 mM ammonium sulfate, 2 mM beta-mercaptoethanol, and 100 mM MES, pH 5.75 in a sealed container containing 500 microlitres reservoir solution, incubated for 3-7 days at 20 degrees Centigrade provide diffraction quality crystals. (473 pages)

- L207 ANSWER 4 OF 14 MEDLINE on STN DUPLICATE 2
- AB Gram-negative bacteria can communicate with each other by N-acyl homoserine lactones (AHLs), which are **quorum-sensing** autoinducers. Recently, the *aiiA* gene (encoding an enzyme catalyzing the degradation of AHL) has been cloned from *Bacillus* sp. strain 240B1. During investigations in the course of the ongoing *Bacillus thuringiensis* subsp. *morrisoni* genome project, an *aiiA* homologue gene in the genome sequence was found. These results led to consideration of the possibility of the widespread existence of the gene in *B. thuringiensis*. *aiiA* homologue genes were found in 16 subspecies of *B. thuringiensis*, and their sequences were determined. Comparison of the *Bacillus* sp. strain 240B1 *aiiA* gene with the *B. thuringiensis* *aiiA* homologue genes showed high homologies of 89 to 95% and 90 to 96% in the nucleotide sequence and deduced amino acid sequence, respectively. Among the subspecies of *B. thuringiensis* having an *aiiA* gene, the subspecies *aizawai*, *galleriae*, *kurstaki*, *kyushuensis*, *ostriniae*, and *subtoxicus* were shown to degrade AHL. It was observed that **recombinant** *Escherichia coli* **producing** *AiiA proteins* also had AHL-degrading activity and could also attenuate the plant pathogenicity of *Erwinia carotovora*. These results indicate that insecticidal *B. thuringiensis* strains might have potential to compete with gram-negative bacteria in natural ecosystems by autoinducer-degrading activity.

- L207 ANSWER 5 OF 14 LIFESCI COPYRIGHT 2006 CSA on STN DUPLICATE 3
- AB *Porphyromonas gingivalis* is a Gram-negative black-pigmented obligate anaerobe implicated in the aetiology of human periodontal disease. The virulence of *P. gingivalis* is associated with the elaboration of the cysteine proteases Arg-gingipain (Rgp) and Lys-gingipain (Kgp), which are produced at high bacterial cell densities. To determine whether **quorum sensing** plays a role in the regulation of Rgp and Kgp, biosensors capable of detecting either N-acylhomoserine lactone (AHLs) or the luxS-dependent autoinducer (AI-2) **quorum-sensing** signalling molecules in spent culture supernatants were first employed. While no AHLs could be detected, the *Vibrio harveyi* BB170 biosensor was activated by spent *P. gingivalis* W50 culture supernatants. The *P. gingivalis* luxS gene was cloned and demonstrated to restore AI-2 production in the *Escherichia coli* luxS mutant DH5 alpha. Mutation of luxS abolished AI-2 **production** in *P. gingivalis*. Western blotting using antibodies raised against the **recombinant protein** revealed that LuxS levels increased throughout growth even though AI-2 activity was only maximally detected at the mid-exponential phase of growth and disappeared by the onset of stationary phase. Similar results were obtained with *E. coli* DH5 alpha transformed with luxS, suggesting that AI-2 production is not limited by a lack of LuxS protein. Analysis of Rgp and Kgp protease activities revealed that the *P. gingivalis* luxS mutant produced around 45% less Rgp and 30% less Kgp activity than the parent strain. In addition, the luxS mutant exhibited a fourfold reduction in haemagglutinin titre. However, these reductions in virulence determinant levels were insufficient to attenuate the luxS mutant in a murine lesion model of *P. gingivalis* infection.

- L207 ANSWER 6 OF 14 MEDLINE on STN DUPLICATE 4
- AB Addition of selected amino acids could be a means to improve

production of recombinant proteins in industrial processes. We found that glycine increased the maximum specific growth rate of *Escherichia coli* from 0.67 to 0.78 h⁻¹, and the cell yield from 0.57 to 0.98 g dry weight per g substrate, when supplemented to batch cultures in a glucose-mineral medium. Maximum effect occurred at pH 6.8, at a glycine concentration of 6-12 mmol l⁻¹, and at cell densities below 1.15 g dry weight l⁻¹ (OD(610).3). When glycine was added to a culture at a cell density of 1.15 g l⁻¹ or above, no growth promoting effect of glycine was seen. The 'glycine effect' was not due to CO₂ produced by the glycine cleavage system (GCV), and the lack of effect at higher cell densities was not masked by acetate accumulation, but coincided with increased acetate production. The metabolism of glycine was further investigated in cultures supplied with [2-(13)C] labelled glycine, and the redistribution of label in the [1-(13)C], [2-(13)C], and [1,2-(13)C] isotopomeres of excreted acetate was analysed by ¹³C NMR. The NMR data revealed that very little degradation of glycine occurred at cell densities below 1.15 g l⁻¹. Simultaneously the biosynthesis of serine and glycine was repressed as judged by the absence of [2-(13)C] acetate, implying that added glycine was used as a source of glycine, serine, one-carbon units, and threonine. At cell densities above 1.15 g l⁻¹, 53% of the consumed glycine carbon was excreted as acetate. Degradation of glycine was associated with an increased uptake rate, cleavage by GCV, and degradation of both glycine-derived serine, and glucose-derived serine to pyruvate. This switch in metabolism appears to be regulated by **quorum sensing**.

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AB Recent evidence has demonstrated that cell-to-cell signaling is a fundamental activity carried out by numerous microorganisms. A number of specialized processes are reported to be regulated by density-dependent signaling molecules including antibiotic production, bioluminescence, biofilm formation, genetic competence, sporulation, swarming motility and virulence. However, a more centralized role for **quorum sensing** is emerging where quorum signaling pathways overlap with stress and starvation circuits to regulate cellular adaptation to changing environmental conditions. The interplay of these phenomena is especially critical in the **expression of recombinant proteins** where elicitation of stress responses can dramatically impact cellular productivity. .COPYRGT. 2002 DeLisa and Bentley; licensee BioMed Central Ltd.

L207 ANSWER 8 OF 14 HCAPLUS COPYRIGHT 2006 ACS on STN

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L207 ANSWER 9 OF 14 MEDLINE on STN

DUPPLICATE 5

AB Numerous gram-negative bacteria employ a cell-to-cell signaling mechanism, termed **quorum sensing**, for controlling gene expression in response to population density. Recently, this phenomenon has been discovered in *Escherichia coli*, and while pathogenic *E. coli* utilize **quorum sensing** to regulate pathogenesis (i.e., expression of virulence genes), the role of **quorum**

sensing in nonpathogenic *E. coli* is less clear, and in particular, there is no information regarding the role of **quorum sensing** during the overexpression of recombinant proteins. The production of autoinducer AI-2, a signaling molecule employed by *E. coli* for intercellular communication, was studied in *E. coli* W3110 chemostat cultures using a *Vibrio harveyi* AI-2 reporter assay (M. G. Surrette and B. L. Bassler, Proc. Natl. Acad. Sci. USA 95:7046-7050, 1998). Chemostat cultures enabled a study of AI-2 regulation through steady-state and transient responses to a variety of environmental stimuli. Results demonstrated that AI-2 levels increased with the steady-state culture growth rate. In addition, AI-2 increased following pulsed addition of glucose, Fe(III), NaCl, and dithiothreitol and decreased following aerobiosis, amino acid starvation, and isopropyl-beta-D-thiogalactopyranoside-induced expression of human interleukin-2 (hIL-2). In general, the AI-2 responses to several perturbations were indicative of a shift in metabolic activity or state of the cells induced by the individual stress. Because of our interest in the **expression of heterologous proteins** in *E. coli*, the transcription of four quorum-regulated genes and 20 stress genes was mapped during the transient response to induced expression of hIL-2. Significant regulatory overlap was revealed among several stress and starvation genes and known **quorum-sensing** genes.

- L207 ANSWER 10 OF 14 MEDLINE on STN DUPLICATE 6
AB Recent reports have shown that bacterial cell-cell communication or **quorum sensing** is quite prevalent in pathogenic *Escherichia coli*, especially at high cell density; however, the role of **quorum sensing** in nonpathogenic *E. coli* is less clear and, in particular, there is no information regarding the role of **quorum sensing** in overexpression of plasmid-encoded genes. In this work, it was found that the activity of a quorum signaling molecule, autoinducer-2 (AI-2), decreased significantly following induction of several plasmid-encoded genes in both low and high-cell-density cultures of *E. coli*. Furthermore, we show that AI-2 signaling level was linearly related to the accumulation level of each protein product and that, in general, the highest rates of **recombinant protein** accumulation resulted in the greatest attenuation of AI-2 signaling. Importantly, our findings demonstrate for the first time that recombinant *E. coli* communicate the stress or burden of overexpressing heterologous genes through the quorum-based AI-2 signaling pathway.
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- L207 ANSWER 11 OF 14 BIOSIS COPYRIGHT (c) 2006 The Thomson Corporation on STN
AB Addition of selected amino acids could be a means to improve production of **recombinant proteins** in industrial processes. We found that glycine increased the maximum specific growth rate of *Escherichia coli* from 0.67 to 0.78 h⁻¹, and the cell yield from 0.57 to 0.98 g dry weight per g substrate, when supplemented to batch cultures in a glucose-mineral medium. Maximum effect occurred at pH 6.8, at a glycine concentration of 6-12 mmol l⁻¹, and at cell densities below 1.15 g dry weight l⁻¹ (0D610cntdot3). When glycine was added to a culture at a cell density of 1.15 g l⁻¹ or above, no growth promoting effect of glycine was seen. The 'glycine effect' was not due to CO₂ produced by the glycine cleavage system (GCV), and the lack of effect at higher cell densities was not masked by acetate accumulation, but coincided with increased acetate production. The metabolism of glycine was further investigated in cultures supplied with (2-¹³C) labelled glycine, and the redistribution of label in the (1-¹³C), (2-¹³C), and (1,2-¹³C) isotopomeres of excreted acetate was analysed by ¹³C NMR. The NMR data revealed that very little degradation of glycine occurred at cell densities below 1.15 g l⁻¹. Simultaneously the biosynthesis of serine and glycine was repressed as judged by the absence of (2-¹³C) acetate, implying that added glycine was used as a source of glycine,

serine, one-carbon units, and threonine. At cell densities above 1.15 g 1-1, 53% of the consumed glycine carbon was excreted as acetate. Degradation of glycine was associated with an increased uptake rate, cleavage by GCV, and degradation of both glycine-derived serine, and glucose-derived serine to pyruvate. This switch in metabolism appears to be regulated by **quorum sensing**.

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AB The *Escherichia coli* mutant CWML2 was previously reported to exhibit improved physiological characteristics, including **recombinant protein production**. Here we investigate the molecular basis of this phenotype by comparing the cellular level of three RNA polymerase sigma subunits by immunoblot analysis. While the level of housekeeping σ .^{sup.D} was similar in parent and mutant, the levels of the flagella synthesis regulator σ .^{sup.F} and the stationary phase regulator σ .^{sup.S} were higher in the mutant strain, indicating a different motility and stationary phase phenotype. Evidence for this conclusion was provided by the significantly higher motility of CWML2, compared to its parent. Based on these results, we hypothesized that alterations in ppGpp regulation via a homoserine lactone-dependent mechanism may be relevant for the mutant phenotype. Indeed, transcription of the *rspAB* operon, which was previously described to be involved in the degradation of homoserine lactone, was found to be deregulated in CWML2 in a plasmid-based reporter protein assay. By overexpression of the *E. coli* *rspAB* operon, we could partly mimic the mutant phenotype and demonstrate that co-overexpression of RspAB is a pertinent metabolic engineering strategy to improve **recombinant protein production**. .COPYRGT. 2000 Academic Press.

L207 ANSWER 13 OF 14 MEDLINE on STN

DUPLICATE 7

AB *Staphylococcus aureus* is a potent human pathogen that expresses a large number of virulence factors in a temporally regulated fashion. Two pleiotropically acting regulatory loci were identified in previous mutational studies. The *agr* locus comprises two operons that express a **quorum-sensing system** from the P2 promoter and a regulatory RNA molecule from the P3 promoter. The *sar* locus encodes a DNA-binding protein that activates the expression of both *agr* operons. We have cloned the *sarA* gene, **expressed SarA in Escherichia coli** and purified the **recombinant protein** to apparent homogeneity. The purified protein was found to be dimeric in the presence and absence of DNA and to consist mostly of alpha-helices. DNase I footprinting of SarA on the putative regulatory region cis to the *agr* promoters revealed three high-affinity binding sites composed of two half-sites each. Quantitative electrophoretic mobility shift assays (EMSA) were used to derive equilibrium binding constants (KD) for the interaction of SarA with these binding sites. An unusual ladder banding pattern was observed in EMSA with a large DNA fragment including all three binding sites. Our data indicate that SarA regulation of the *agr* operons involves binding to multiple half-sites and may involve other sites located downstream of the promoters.

L207 ANSWER 14 OF 14 MEDLINE on STN

DUPLICATE 8

AB The *enterobacterium* *Erwinia carotovora* ssp. *carotovora* strain 71 (hereafter Ecc71) produces extracellular enzymes such as pectate lyase isozymes (Pels), cellulase (Cel), polygalacturonase (Peh) and protease (Prt). These enzymes degrade plant cell wall components and are largely responsible for the elicitation of soft-rot diseases in plants and plant products. Ecc71 also produces HarpinEcc, the elicitor of hypersensitive reaction (HR) and the **quorum-sensing signal**, N-(3-oxohexanoyl)-L-homoserine lactone (OHL). OHL controls extracellular enzyme and HarpinEcc production. The levels of these enzymes, as well as the expression of *hrpNEcc*, the structural gene for HarpinEcc, and *ohl1*, the gene specifying OHL synthesis, are negatively regulated by *RsmaA*. *rsmB*, formerly *aepH*, on the other hand, positively regulates extracellular

enzyme production. 6His-RsmA recombinant protein purified from E. coli binds rsmB RNA as indicated by gel mobility shift assays. rsmB comprises 547 bp DNA, which is transcribed from a single start site immediately after a sigma70-like promoter. In Ecc71, two rsmB RNA species are detected: a full-length 479 base rsmB RNA and a 259 base rsmB' RNA. rsmB' DNA hybridizes with the 259 base and the 479 base transcripts. A 3' RNase protection assay revealed that the 259 base and the 479 base RNA species end at the same position immediately after the putative rho-independent terminator. The expression of rsmB-lacZ transcriptional fusions established that the rsmB' RNA is not produced because of the activation of an internal promoter. These data strongly suggest that the 259 base rsmB' RNA is derived by processing of the primary rsmB RNA. In Ecc71, rsmB' expression driven by the lac promoter causes overproduction of Pel, Peh, Cel and Prt, and accumulation of pel-1, peh-1, hrpNEcc and ohll transcripts. By contrast, a plasmid with the rsmB' DNA sequence deleted fails to cause overproduction of the extracellular enzymes in Ecc71. The rsmB' effect also occurs in Escherichia coli as glycogen accumulation is stimulated in the presence of rsmB'. In vivo and in vitro translation as well as mutational analysis of rsmB' have established that rsmB' RNA does not yield a translational product. Therefore, we concluded that the rsmB' RNA itself functions as the regulator. Indeed, the expression rsmB' DNA leads to neutralization of the negative effects of the RNA-binding protein, RsmA, in Ecc71 and Serratia marcescens strain SM274. We propose a model that explains how RsmA and rsmB control the expression of genes for extracellular enzymes.

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